THE PROPIONIC ACID TREATMENT OF MOIST HAY

David Lindsay Easson

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DECLARATION

This thesis has been composed by myself and describes experimental work which I carried out.

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SUMMARY

The aim was to investigate the use of propionic acid as a preservative for moist hay. In two field experiments propionic acid was applied using a high or a low pressure sprayer attached to the baler, to hay at moisture contents between 24 and 47 per cent. Gross application rates of up to 5 per cent by weight were unable to prevent mouldy patches appearing in bales with over 30 per cent moisture. Treatments of between 1 and 5 per cent increasingly delayed and reduced the moulding, heating and losses. At below 28 per cent moisture, levels of acid of 2 per cent and above prevented visible moulding when applied using the high pressure applicator, but 5 per cent acid was needed when the low pressure applicator was used in the second experiment. The pattern of changes taking place in 9 bale batches was similar to that in 50 bale batches.

Experiments with miniature bales containing between 1kg and 2kg of hay treated uniformly with propionic acid resulted in the preservation of hay at approximately 30, 40 and 50 per cent moisture by acid levels of 1.0, 1.5 and 2.0 per cent acid respectively. In higher moisture material, including fresh grass, under aerobic conditions, acid at between 3 and 7.5 per cent provided preservation although some unexplained loss of water soluble carbohydrate did take place. The minimum acid levels for preservation fitted the equation:-

(1) Acid level =(Moisture content - 15) x 0.06 per cent.

The variations in moisture content and propionic acid distribution were measured in bales made from experimental swaths representing the best and worst of possible conditions, treated with

acid using two designs of low pressure applicator. Neither was able to give an even distribution, the parts of the bales containing hay which passed near the spray jets having higher acid levels. The levels on different parts within some bales had a range of more than 10 per cent acid. If the profile of the swath at baling was uneven the variations in acid distribution within the bales became greater, but even in bales from the most uniform swaths considerable variation occurred. The moisture contents also had a wide range but the variations in acid were the main cause of mouldy patches in acid treated bales.

The estimates of moulding using equation (1) were too low for conditions in which the acid distribution was not uniform. It was estimated that under field conditions with the present applicators net acid levels of 6 to 7 per cent by weight on the hay would prevent moulding, while 4.5 to 6 per cent would confine moulding to 3 per cent of hay baled at between 25 and 30 per cent moisture.

It was suggested that, if economic, propionic acid should at present only be used to reduce moulding and losses in hay which is unavoidably being baled moist, or as a safeguard on hay baled at the normal time, but not in an attempt to preserve moist hay which could otherwise be dried satisfactorily in the field or barn.

INTRODUCTION

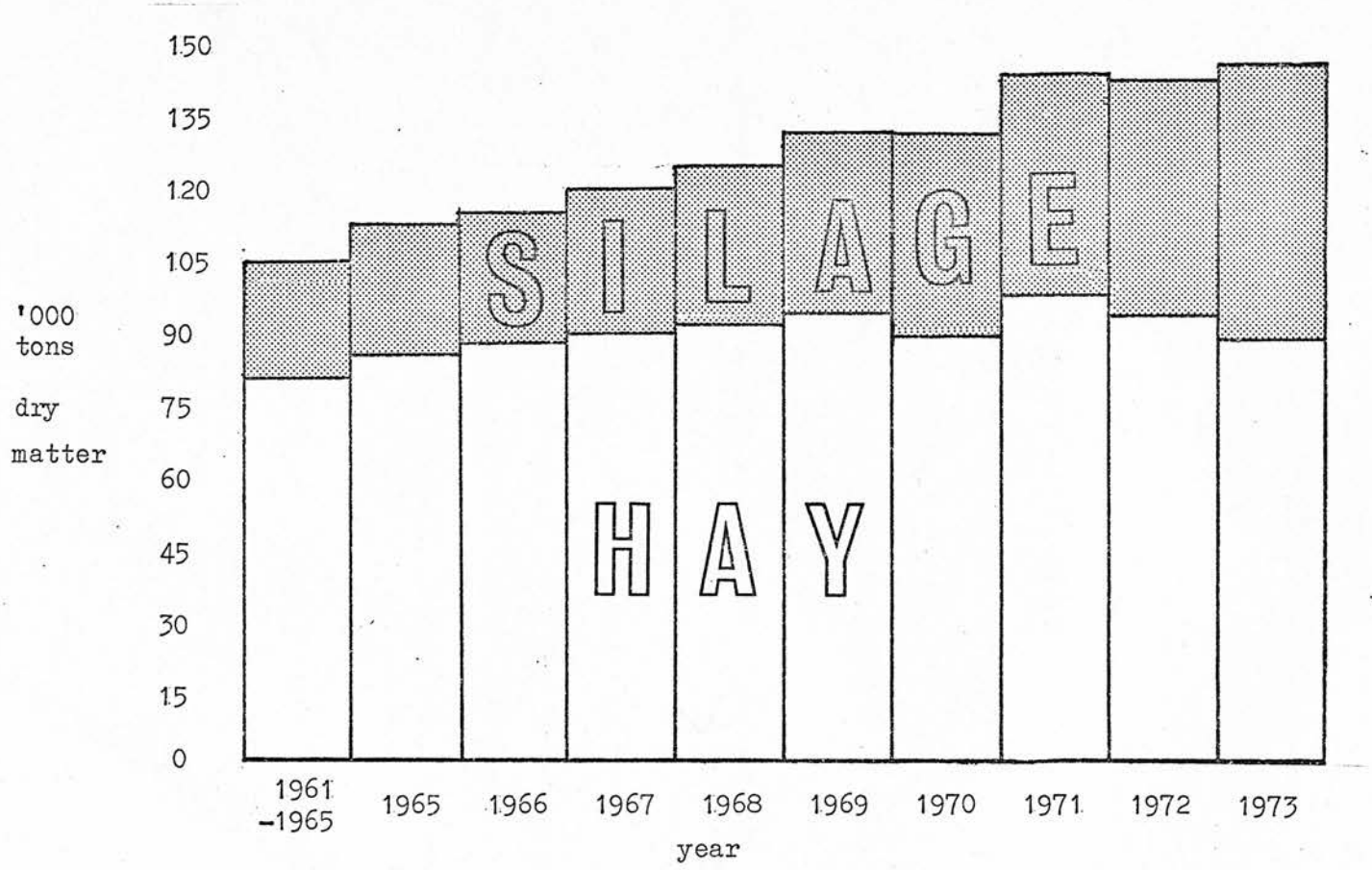
The term 'hay' is familiar to all but the most urbanised of our population. It refers to the product of one method by which a farmer may conserve some of the excess herbage production of the summer and autumn for feeding to his stock in the winter. The conservation of herbage destined for hay has traditionally been achieved by natural drying in the field, before storage. For this method to be effective the moisture content must be reduced to below the level at which plant cells will respire and the agents of bio-deterioration, principally fungi, actively proliferate. This low moisture content must be maintained throughout the storage period.

In current farming practice it is sometimes the case that only part of the drying process is achieved in the field, the final stages of drying being carried out by means of a 'barn-drying' system after the hay has been brought into storage. If haymaking is to be completed in the field, however, the final and most difficult drying stages, during which severe losses can easily occur, are commonly carried out with the hay in small stacks of bales or loose hay which afford some protection against the deleterious effects of rain and sun.

As the traditional form of conservation for fodder in the United Kingdom, hay has played a leading role in the winter rations of all classes of ruminant livestock. However, in recent years there has been a greater realisation of the importance of conserving herbage near to its peak nutritional value. Thus in systems which demand more intensive feeding methods, such as dairying, silage as an alternative to hay has gained in popularity (Figure I), since it is capable of being easily made at an eariler and more digestible stage of growth and without the same dependence upon good weather conditions.

Figure I

Hay and Silage Production in Scotland, 1961 to 1973



Source: Department of Agriculture and Fisheries for Scotland (1968 - 1974).

The quantity of grass conserved as hay, although showing a slight downward trend in the last few years, is still considerably greater than that conserved as silage, and in terms of dry matter constituted over 60 per cent of the total in 1973. Valued at the current price of about £35 per ton (September, 1975) the hay crop in Scotland is worth over £38.7 million.

For many years mechanical techniques have been available to the farmer by which, under reasonable weather conditions, he can ensure rapid field drying of his crop. Reasonable weather, however, is not a consistent feature of our climate, and the probability of the weather forecast predicting the ideal 3 day period of warm dry weather is not high (Hughes, 1967). Whether from this cause, or due more directly to inadequate management, hay is often baled and brought into storage before the moisture content has been sufficiently reduced. As a result the hay frequently undergoes moulding accompanied by heating and the consequent loss of nutritive value, followed on rare occasions by complete loss of the hay due to spontaneous combustion.

Farmers, however, have always been reluctant to invest capital in an improved haymaking system because the crop has been regarded as a maintainance roughage which shows little return on investment. Barn--drying systems, for example, have not been widely adopted even although this method has been shown to be economically justifiable in terms of improved recovery of dry matter (Shepperson, 1971).

The obvious need for a low capital cost system for improving hay quality by reducing the dependance of haymaking upon good weather and preventing losses due to moulding has stimulated the appearance on the market in recent years of a number of chemical additives for hay. If such additives were to prove to be effective and economic then there is little doubt that they would be widely adopted. Apart from

their obvious scope for bringing about a general improvement in the quality of hay effective additives could be of particular importance in reducing the incidence of farmer's lung disease attributable to mouldy hay. The incidence of this disease is particularly high among the farming populations of Wales and the North and West of Scotland (Grant et al, 1972).

In most, if not all, of the commercial additives available at present propionic acid is the principal ingredient capable of inhibiting the growth of moulds. In some of the additives, however, the percentage of propionic acid is low and in trials the additives have usually failed to bring about any significant improvement in quality when used at the rates recommended by the manufacturers (A.D.A.S., 1968 - 1973). The additives have also been shown to be less effective than pure propionic acid used under the same conditions.

For many years propionic acid and its derivatives have been used as commercial preservatives in sliced bread and other food products, and since 1968 the pure acid has been marketed as a preservative for moist grain. Its success for this type of farm storage prompted interest in its use in a similar way to preserve hay. The acid is manufactured by BP Chemicals (International) Ltd., as a by-product of their petro-chemical division, and it was through their financial support that the experimental work described in this thesis was carried out.

The emphasis throughout the work has been to investigate the use of propionic acid as a preservative for moist hay within the normal systems of haymaking, although some of the work is bound to have wider implications. No attempt has been made to evaluate the results in economic terms, but it must be acknowledged that at the end of the day it may well be the economic factors which dictate whether such a method of preservation of hay is feasible or not.

REVIEW

Propionic acid is a short chain member of the fatty acid series which, as a whole, is recognised to have antibiotic activities. Investigations with these acids have been recorded since 1896 and the work of Kiesel (1913) at the Pastew Institute has provided much basic information on their antimycotic action. He established that the activity of the saturated acids increases with the number of carbon atoms in the chain up to 12 atoms, but that branched chains acids are less active than the corresponding straight chain acids. Kiesel also established that substitution of hydrogen ions decreases activity, while in Japan Tetsumoto (1933) found unsaturated acids to be more active than the corresponding saturated acid, and decarboxylic acids to be almost without activity. In Germany Wyss et al (1945) compared propionic acid with many other longer chain saturated and unsaturated acids. Propionic acid was the least inhibitory to the growth of moulds. In addition its effect was more easily reduced by a rise in pH and consequent changes in ion concentration. These workers found that solubility was the factor which limited the activity with the length of the carbon chain. In considering the mycocidal as opposed to inhibitory effect of the acids Wyss et al concluded that the killing of the cell is merely a more drastic expression of the disturbance, which is the same or very similar to that producing inhibition.

The undissociated acid molecule was thought by Kahlenberg and True (1896) in the USA, to be the active agent, as it was clear that the fatty acids exerted a mycostatic power greater than the hydrogen ion concentration alone would provide (Paul et al., 1910). Tetsumoto (1933) and Strider and

Winsted (1960) found the dissociated acid anions to be non toxic and also concluded that the undissociated molecules were mainly responsible for the inhibitory effects of these acids. It was suggested by Lewis (1951) in the USA that other factors such as surface tension and the molecular weight and size of the molecule might also be of importance, and that the overall effect should more correctly be thought of as a combination of factors. The work of Kirby et al (1937), Hoffman et al (1940) and Rigler and Greathouse (1940) served to confirm these basic conclusions and establish them over a range of conditions and organisms.

With the current increase of interest in the use of propionic as a commercial preservative of grain and hay a number of papers have been published over the last few years which have shed further light on the antimycotic action of both propionic acid and the other short chain fatty acids. Comprehensive laboratory tests were carried out by Woolford (1975) on the inhibitory effects of the C_1 to C_{12} straight chain fatty acids on the microorganisms found in silage. These included a number of fungi, such as Aspergillus fumigatus, which are also commonly found on mouldy hay. This work showed once again the increasing mycostatic action of the fatty acids as the carbon chain lengthens, up to 9 carbon atoms (pelargonic acid), but the increases in mycostatic action above butyric acid (4 carbon atoms) were only slight. The action of the acids was also shown to increase as the pH became lower. Propionic acid was about 4 times as active at pH 5 as at pH 6, while butyric acid was 8 times as active. When the pH values were reduced to 4 or 3, however, increases were slight. Woolford concluded that when acting as an inhibitor the effect of a fatty acid would be due to a combination of the effect of the concentration of hydrogen ions (pH)

together with the action of the acid molecule itself. This supports the conclusions of earlier workers. However, he did not consider his results to provide any information as to whether the active agent of the acid was the dissociated ion or undissociated molecule. An explanation of the significance of the hydrogen ion concentration above is given by Simon and Blackman (1949).

As a result of experiments on the preservation of high moisture maize (20 per cent moisture content) with various short chain fatty acids Herting and Drury (1974) found that the simple branch chain acid, isobutyric acid, was more active as a mycostatic agent than formic, acetic, propionic or butyric acids. This runs counter to the earlier conclusion of Kiesel (1913), but as this acid is not quoted in other work there is no way of confirming their conclusion. Herting and Drury also found that mixtures of the fatty acids were more active than single acids, the most effective of the mixtures they tried containing propionic and isobutyric acids in equal proportions, but the workers were unable to provide an explanation for this phenomenon. In a second series of experiments by Herting et al (1974), aqueous solutions of formic, acetic, propionic, butyric and isobutyric acids were tested as mycostatic agents. It was found that in dilutions containing over 50 per cent water the fungicidal activity of the acid on the maize increased rather than decreased. For example, using a mixture of propionic, butyric and isobutyric acids (50:25:25) the quantity of acid required to prevent fungal activity was halved when the acid was applied in a solution with 90 per cent water. At concentrations below 50 per cent water, however, the mycostatic action of the acid was unaltered and the application of the solution would have the same result as increasing the moisture content of the substrate.

The workers suggested that the reason for the increased activity in relatively dilute solutions could be that the excess water could split the hydrogen bonds which normally holds the fatty acid molecules in pairs (dimers) to form new dimers of water plus the fatty acid. In this way the number of potentially active molecules would be doubled.

Few workers have considered where the site of action of the acids on the cell might be. Strider and Winsted (1960) found that a site within the cell was indicated for some species while results with others favoured a site on the cell surface. Fencl and Leopold (1957) concluded that with the action of acetic acid on Aspergillus niger the effect was an internal cellular one. If, as is generally accepted, the undissociated molecule is the main inhibiting agent it could be that the site of action is within the cell, because a neutral molecule would find it easier than a charged ion to permeate through the cell wall (Byrde, 1969). Neiman(1954) suggested that if the acid entered the cells it could affect the cell wall permeability and either block the absorption of essential nutrients thus inhibiting the organism, or cause outward diffusion of vital cell components resulting in the death of the cell.

The mycostatic effect of the fatty acids, including propionic acid, is also possessed by various salts of the acids. Calcium propionate, for example, has been widely used as a preservative to increase the shelf life of sliced bread and other foods (Lamprecht, 1955). Halick and Richardson (1953) demonstrated that calcium propionate could effectively inhibit moulding on grain, while a few years later they tested a wider range of derivatives including sodium propionate, propionamide propionanalide and propionic acid itself (Richardson and Halick, 1957). Other than propionic

acid, calcium propionate was the most effective inhibitor of mould growth. Earlier work by Wolford (1945) had shown sodium propionate to be an effective inhibitor of bacteria, while at the present time ammonium propionate is being tested as a hay preservative (Mandy and Shepperson, 1975).

Propionic acid, however, is not equally inhibitory to all species of mould, and it has been shown by Lacey and Lord (1974 a & b) that at least one species of mould, Paecilomyces varioti, is able to metabolise low levels of propionic acid as an energy source and therefore degrade the acid. Higher levels of acid did inhibit this species of mould which, in earlier work, had emerged as being particularly resistant to propionic acid inhibition (Lacey and Hill, 1969). Although there is no evidence of it at present, there is always the risk that propionic acid resistant strains of moulds will develop more widely and make propionic acid less reliable as a preservative.

The action of the fatty acids is not just against moulds, but has a wide spectrum against many species of bacteria and other organisms. Woolford (1975) demonstrated their activity against lactic acid and spore-forming bacteria as well as moulds and yeasts found in silage, while other work has shown their effectiveness against pathogenic fungi (Thornton, 1963), pathogenic bacteria (Dubos, 1950) and food spoilage insects (Burkholder et al., 1973).

Propionic acid also causes the rapid death of plant cells and when applied to grain kills the embryos and so prevents the germination of the seeds (Sauer et al., 1972). The cells of grass treated with propionic acid are plasmolysed so that the cell fluids readily escape. This property

has recently been used to speed the desiccation of grass cut for making wilted silage or hay (Pirkelmann, 1972; Wieneke and Hartmann, 1971). However, propionic acid does not seem to be as effective as formic acid in this context and after a day or two of drying the acids are almost entirely lost so that their anti-microbial action in subsequent stored crops would be minimal.

Propionic Acid Preservation of Hay

Research work into the use of fatty acids as potential preservatives for farm crops is not extensive, but such as has been done is mainly concerned with the short chain acids such as propionic acid. The longer chain acids, although more inhibitory, have not been considered due to their high cost and their physical properties which make them more difficult to apply as a preservative. The longer chain acids are solids of low solubility, whereas propionic acid is a liquid, completely miscible with water.

The first experiments on the preservation of hay seem to be those of Lewis (1951) in America. In his work he first of all isolated the moulds most commonly growing on hay and found them to be mainly of Mucor, Aspergillus and Penicillium species. In laboratory experiments he then tested the growth of moulds of these species on media containing valeric, butyric, propionic and acetic acids at various concentrations. In common with the other workers already referred to he found that the mycostatic activity increased with the length of the carbon chain, with the exception that valeric acid was less active due to its low solubility. Lewis continued his investigation by spraying 300 gram portions of hay at 30 to 40 per cent moisture with the acids, using a paint spray gun to provide even application. The treated hays were stored in glass jars in which any moulding could be easily seen.

Butyric acid at levels above 1 per cent by weight, and propionic acid at above 2 per cent both preserved the hay over a three month period. Valeric acid, however, required an application rate of at least 4 per cent to be effective over this period, while even at a 5 per cent application rate acetic acid could not inhibit moulding for more than a few days. Lewis observed that, although moulding might be delayed by an acid treatment, once a patch of mould did develop its spread throughout the hay in that jar was rapid and inevitable.

Although butyric acid was the most effective of the acids he tried, Lewis suggested that the strong odour of this acid would make it unacceptable for practical use and so he went on to try field experiments with propionic acid. These, however, were a total failure as he was unable to find a suitable method of applying the acid to the hay. He tried spraying the grass at the time of cutting, or the swath prior to baling, but found that in both cases the acid volatilised from the hay before baling took place. He therefore concluded, as many have done since, that the application of the acid to the hay is the main stumbling block to its use on the farm as a hay additive.

Problems during the treatment of hay with mould inhibitors in the field were also encountered by Kennedy and Schenk (1954). While experimenting with trichlorophenol (TCP) and dichlorobenzene (DCB), which proved to be effective mould inhibitors on hay in lab. experiments (Dawson et al., 1950), they found that application of the chemicals at the time of cutting or raking could lead to high losses due to leaching or evaporation while application at the time of baling made it difficult to apply the chemicals evenly. They also observed that, regardless of method of application, the

leaves which make up most of the surface area receive most of the treatment, while the stems which are likely to contain more moisture receive less of the treatment.

Hopkins and Wiant (1956) failed in their attempt to find a suitable mould inhibitor for hay, but did not test propionic acid. From their experiments however, they suggested that the best distribution of a liquid fungicide could be obtained by using a 2 or 3 nozzle spray-boom mounted above the baler apron with a hood to reduce spray drift in the wind.

Interest in the use of propionic acid and in hay preservatives in general lapsed until in 1968, after 3 years of their own investigations, BP Chemicals (International) Ltd., marketed propionic acid as a commercial preservative for grain. Trials carried out at Experimental Husbandry Farms (National Agricultural Advisory Service 1968, 1969 and 1970) and at the Edinburgh School of Agriculture (Nash, Wallace and Whittenbury, 1968; Nash, Easson, Whittenbury and Robb, 1969), established that at the rates of propionic acid recommended by the manufacturers (BP Chemicals (International) Ltd., 1968) propionic acid could effectively preserve grain over a range of moisture contents. Successful preservation of shelled maize (Jones, 1970) sorghum and wheat (Sauer et al., 1972), ground maize (Goering and Gordon, 1973) and field beans (Nash, Hodgson-Jones and Easson, 1970) at various moisture contents have since demonstrated the effectiveness of propionic acid in the prevention of moulding in high moisture grain crops. No ill effects have been reported when propionic acid treated grain was fed to dairy cattle or pigs (Jones et al., 1970; Young, et al., 1970; Whittemore and Taylor, 1973).

With the success of the propionic acid treatment of grain crops attention turned to the possibility of preserving moist hay with the same chemical. It was at that time, in 1969, that the investigations reported in this thesis were begun at the Edinburgh School of Agriculture. Interest in this use of propionic acid also led to a series of laboratory and field experiments by Lacey at Rothamsted and by Shukking at Wageningen in Holland, and to field trials at Drayton, Liscombe and other Experimental Husbandry Farms of the Agricultural Development and Advisory Service.

Although it is possible to piece together the information from these different centres, the work is mostly in the form of trials without replication or statistical analyses of the results and therefore it is difficult to draw any definite conclusions. The task is made more difficult by the variety of methods employed for applying the propionic acid in the field and by the different types of hay tested in the laboratory.

Lacey (1968 and 1974) found that hay which was rewetted and stored in vacuum, (dewar) flasks after treatment with propionic acid, could be preserved at lower rates of acid than those suggested by Lewis (1951) or Shukking (1972). Less than one half per cent of propionic acid preserved hay of 30 per cent moisture, while 1.0 per cent preserved hay of 50 per cent moisture in Lacey's work, while the other workers who used freshly made hay found that 2.0 and 2.5 to 3.0 per cent acid respectively were required at these moisture levels. Lacey (1968) also found that propionic acid was more effective than formic acid while in contrast Morgan-Jones (1969) whose results were on the whole inconclusive, considered formic acid to have been more inhibitory than propionic acid when tested on 10g samples of damp hay. Goering and Gordon (1973), working in the U.S.A., used ground

and rewetted alfalfa and cocksfoot hays stored in small (0.25 litres) glass jars and found results similar to those of Lacey in that 1.0 per cent propionic acid was sufficient to preserve hay at 50 to 55 per cent moisture.

The nature of the hay sample and its method of storage has obviously had a significant effect upon the results of these experiments. If one is trying to establish the net acid levels which would be required to preserve hay made and treated in the field the results of Lacey's and Goering and Gordon's work in which rewetted material was used, cannot be taken as a reliable guide. Shukking (1972) used the method of storing the fresh treated hay in insulated boxes 28 x 22 x 26 cm which probably gives the most reliable comparison with normal hay storage conditions, but he gives few details of the design and results of the experiments upon which he bases his conclusions that net acid levels of 1.0, 1.5 and 2.0 per cent are needed to preserve hay at moisture contents of 30, 35 and 40 per cent respectively.

The results of the field experiments which have been carried out are summarised on the next two pages. Taking the results as a whole, it is clear that moulding was not prevented in any of the trials in which hay, with a moisture content of above 33 per cent was treated with propionic acid, irrespective of the method of application. Although the results of Candlish et al (1973) in Canada are an exception to this statement they used a mixture of propionic acid and acetic acid on the high moisture hay and not propionic acid alone. The levels of acid found by Candlish et al to satisfactorily preserve hay at the different moisture levels were so much lower than those of all the other workers, even in laboratory experiments, that it is difficult to say how much credence should be given to their results.

Summary of Field Trials with Propionic Acid

1. Experimental Husbandry Farms

EHF	Percentage Moisture Content	Percentage Gross Acid Level	Applicator	Result*
Liscombe 1969	44.8	2.0		Severe moulding
Liscombe 1973	41.0	0.9	F	Severe moulding
Liscombe 1971	35 to 39	1.2	D	Moulding
Drayton 1972	38 to 40	3.6	G	Moulding
Liscombe 1970	36.3	0.7	C	Moulding
Drayton 1972	33.5	3.6	E	Moulding
Liscombe 1973	33.0	0.5	F	Moulding
Liscombe 1971	31 to 33	3.9	D	Satisfactory
Liscombe 1969	32.1	1.5		Satisfactory
Drayton 1968	30	2.5	A	Satisfactory
Drayton 1969	30	2.3	B	Slight moulding
Liscombe 1973	28 to 30	1.8	F	Slight moulding
Drayton 1971	28 to 30	1.9	D	Satisfactory
Drayton 1971	28.9	2.3	D	Moulding
Drayton 1973	28.5	3.6	F	Satisfactory
Drayton 1973	28.5	1.8	F	Satisfactory
Liscombe 1973	23 to 27	0.9	F	Moulding

* Based on the description of the results given in the reports.

'Satisfactory' does not necessarily imply the complete absence of moulding.

A Oscillating single fan jet

B Oscillating multiple fan jet

C Applicator attached to forage harvester

D Two fan jets above baler apron (Low Pressure, Mark I, see Appendix 7)

E Two fan jets across bale chamber inlet (Low pressure, Mark II)

F Jet onto spinning disc above baler apron

G 4 fan jets above baler apron

Summary of Field Trials with Propionic Acid (Cont.)

2. Lacey and Lord (1972)
(Rothamsted)

Percentage Moisture Content	Percentage Acid Level	Applicator	Result
41	2.0*	E	Moulding
28	3.0*	E	Slight moulding
26	3.0*	E	Slight moulding

3. Devuyst et al (1972)
(Belgium)

35	0.75*	at baler	Moulding

4. Shukking (1972)
(Netherlands)

30	1.0@	C	Satisfactory
30	0.6 to 0.7@	C	Satisfactory
35 to 38	0.9@	D	Moulding
25 to 29	1 to 1.5@	D	Satisfactory

5. Candlish et al (1973)
(Canada)

40	0.1+	H	Satisfactory
40	0.07+	H	Satisfactory
26	0.07	H	Satisfactory
26	0.10	H	Satisfactory
18	0.10	H	Satisfactory

* Gross acid level applied

@ Net acid level received by the hay

\+ Treatment with mixture of propionic and acetic acids 3:7 ratio

H Single jet above baler apron

Although the acid treatments did not entirely prevent moulding in the hay with more than 33 per cent moisture they did, in most cases, delay and reduce the heating in the bales and restrict the moulding to patches. Acid treatments of the hays at below 33 per cent moisture tended to be more successful, but not universally so even at the higher levels of acid application above 2 per cent. Lacey and Lord (1972) reported growth of Aspergillus glaucus producing large numbers of yellow cleistothecia on the 28 and 26 per cent moisture hays, even with a 3 per cent treatment level. They suggested that uneven distribution of the acid resulted in these mouldy areas. Shukking considered that application via the forage harvester gave the best distribution, but this resulted in losses of acid of over 50 per cent. From A.D.A.S. (1972) results the applicator with the nozzles at the bale chamber inlet was thought to give a better distribution than the one with the jets above the baler apron, but problems were still being encountered even with the most up-to-date design of applicator in which a fine mist of acid was spread across the baler inlet from a spinning disc onto which a jet of acid played. (A.D.A.S., 1973)

Lacey has investigated the significance of the variations in acid distribution in a number of laboratory experiments. The introduction of layers of untreated moist hay into dewar flasks containing otherwise well preserved propionic acid treated hay resulted in the spread of moulding from the untreated into the treated hay (Lacey and Hill, 1969). This took place when the treated layer received twice the level of acid which would have preserved all the hay had it been applied uniformly, but when the treated layers received four times the adequate level moulding was restricted to the edge of the treated layers. Only when the acid treatment was increased to ten times the adequate level was the influence of the preservative

able to spread from the treated into the untreated areas. Later experimental work (Lacey and Lord, 1972) indicated that, one hour after treatment with propionic acid, the hay had absorbed the acid to the extent that redistribution or loss of the acid in air currents was unlikely to occur to any significant extent. The analysis of individual grass stems from bales of hay made and treated with propionic acid in the field (Lacey et al., 1973) revealed wide fluctuation in acid distribution even along a single grass stem, with similar variations in mould growth. Evidence was found for the spread of mould growth, from areas which received a low acid treatment adjacent to areas with a higher acid level. One species of mould was found which was able to metabolise the acid (Lacey and Lord, 1974) in areas which had received a marginal level of acid treatment (Lacey and Hill, 1969). The degradation of the propionic acid by moulds may have been the source of propionic acid losses recorded during storage by Lacey et al (1973) and A.D.A.S. (1973).

The loss of acid during the treatment of the hay was another source of loss and was repeatedly mentioned by the experimental workers. Shukking (1972) found that losses of between 35 and 55 per cent of the acid took place during treatment and A.D.A.S. (1973) on one occasion recorded that 65 per cent of the acid was lost. Goering and Gordon (1973) found that addition of the acid as chopped hay was being blown into experimental wire framed stacks was not particularly successful and suggested that movement of air through the open 'snow fence' walls of the stacks could have resulted in high losses of the volatile acids. Contrary to the results of their laboratory experiments Goering and Gordon found that the propionic acid, acetic acid mixture (3:7 ratio) treatment at 2.1 per cent and ammonium butyrate treatment at 1.1 per cent were more effective in controlling

heating in these small stacks than the propionic acid treatment of 1.5 per cent. In general, however, propionic acid has been the most successful of the various hay additives compared in field trials and laboratory experiments (A.D.A.S. 1968, 1969, 1970; Lacey and Hill, 1969).

In addition to its use on hay, propionic acid has been used successfully to preserve dried grass wafers stored at high humidity (Chaplin and Tetlow, 1971) and high dry matter silage (Schubking and Hengeveld, 1972; Papendick and Singh-verma, 1972). Shukking (1972) found that the stability was increased, and heating reduced, in silages with dry matters from 35 to 60 per cent treated with net propionic acid levels of between 0.3 and 0.6 per cent which confirmed the earlier work of Daniel et al (1970), in which acid treatments of 0.3 and 1.0 per cent had reduced 'after fermentation' in silages with dry matter levels of 30, 44 and 50 per cent.

Although a number of trials have been carried out on the feeding of propionic acid treated grain to farm animals (Jones et al., 1970; Young, et al., 1970; Whittemore et al., 1973), little work has been carried out on the feeding of acid treated hay. Candlish et al (1973) reported no harmful effects to animals fed acid treated hay. Although the propionic acid treated hay was found to have a lower dry matter digestibility and give a lower intake than the untreated hay it is not clear whether propionic acid treatment of this hay had successfully prevented moulding or not. In the work of Mansour and Jackson (1973), at the Edinburgh School of Agriculture, organic matter digestibility was found to be slightly improved by a propionic acid treatment of 4 per cent, which eliminated virtually all moulding. Lambs fed ad lib on this hay with a barley supplement had a similar growth

rate to those fed conventional hay in the same experiment. Considering that propionic acid is a natural product of the fermentation of roughages in the ruminants it is not surprising that the addition of relatively small quantities of the acid to the diet has not been shown to have any deleterious effect. Jones et al (1970), however, have suggested that in the diet of dairy cattle propionic acid may increase milk protein synthesis, but supress milk fat production.

The weight of evidence suggests that propionic acid has the potential to be an effective hay preservative, and be entirely safe in animal feeding stuffs, but that a method of providing an even distribution of the acid has yet to be devised. The only other additive which has been tested and might provide a similar measure of mould inhibition is a mixture of 3 parts propionic acid with 7 parts acetic acid. From the review, however, the lack is seen of a comprehensive piece of research in which the effect of various acid levels on hay of different moisture contents are compared in a single experiment and in which the factors preventing the satisfactory distribution of the acid could be investigated. The need also remains for more detailed experimental work to establish the net acid levels, under conditions in which the acid is evenly applied, which would effectively preserve hay at different moisture levels. In order to be of value the work would have to be carried out under conditions resembling as far as possible, those of normal hay making. It was with these needs in mind that the research work described in this thesis was undertaken.

The Field Drying, Storage and Deterioration of Hay

Such topics as the drying of hay in the field, respiration of hay in the field and in the store and the relationship between moisture content

and moulding, have a direct bearing on the experiments in this thesis, and so a review of the current literature on these topics is now given. Watson and Nash (1960) have provided a comprehensive cover of the relevant literature up to 1959, and so the present review is intended to cover the most recent literature, with reference to earlier papers only when they are of particular importance.

Field Drying of Hay

The rate of drying of cut herbage is controlled principally by the weather (Eruck and Elderen, 1969; Shepherd, 1965) and the physical condition of the crop (Mitchell and Shepperson, 1955; Shepperson et al., 1962). Loss of moisture may take place very rapidly in the initial stages of drying during suitable weather conditions especially if the crop has received a mechanical treatment, such as crushing or crimping, and if the swath is being turned and tedded frequently. Various other treatments have been developed recently to speed up the rate of drying. Steam treatment of the standing crop before cutting has been developed in Holland (Philipsen, 1969), while the use of chemical desiccants such as formic and propionic acids has also been the subject of experiments in Germany (Pirkelmann, 1972). As the moisture content of the herbage falls, the rate of respiration of the living plant cells declines (Simpson, 1961) but dry matter losses, mainly of soluble carbohydrate (Melvin and Simpson, 1963), in the order of 5 to 10 per cent may still be expected from this source (Watson and Nash, 1960 a) If the drying is prolonged due to poor weather, increased dry matter losses may be expected. Rainfall may further increase the losses by the leaching of soluble fractions of the hay, particularly during the later stages of drying, when most of the plant cells have died and can readily loose their

contents (Watson and Nash, 1960 b).

Moisture is lost rapidly from the outer cells of the leaves and stem and this is reflected in the comparative ease with which the moisture content may be reduced from above 75 per cent at cutting to about 50 per cent. However, as further moisture loss is dependent upon internal diffusion of the moisture to the surface tissues the rate of drying is considerably reduced (Simpson, 1961). The stems take longer to dry than the leaves and so mechanical treatments aimed at cracking the stems are often employed to even out and speed up the drying process. Unfortunately, the more violent the mechanical treatment the greater are the risks of high dry matter losses (Murdoch and Bare, 1963; Shepperson et al., 1962), particularly of the leaf which becomes brittle as the moisture content of the hay approaches 30 per cent, (Zink, 1936). Losses of over 20 per cent of the dry matter may be incurred, particularly with leguminous crops, if there is excessive handling of the dry hay. Physical losses are the more serious because, consisting mainly of leaf, it is the more nutritious and digestible portion of the grass which is lost (Watson and Nash, 1960 C).

A combination of tedding and turning the hay will promote even drying in the swath, but turning alone will not produce this (Murdoch and Bare, 1963). The condition of the swath after the pass of the machine has an important effect on the rate of drying (Shepperson and Grundey, 1962), a fluffy swath drying more rapidly. Heavy rain on bruised material which has been partly dried may flatten the swath and result in the wet patches noted by Mitchell and Shepperson (1955). Such patches are difficult to dry and may eventually ferment and rot. By retarding the removal of the crop from the field the wet patches contribute to the loss of overdried

leaf, but if the crop is removed to save leaf the wet patches become centres of hot spots and reduce the quality of the bales in store. A new crop conditioner is being developed, however, which in preliminary trials has been able to increase the rate of drying to a similar extent as crimping, but retaining the structural strength of the hay so that loose swaths can be formed and rapid and even drying of the swath promoted (Klinner, 1975).

It has been shown by Nash (1962) that under good weather conditions in S.E. Scotland and where the hay is crimped and frequently tedded it may be dried to below 30 per cent moisture within 3 days. Continuous good weather such as this is not a reliable feature of our climate, however, and Hughes (1967) after studying the weather conditions in Wales over a ten year period concluded that, in most cases, there would have been no possibility of reducing the length of time some hay would have had to lie at the mercy of the weather. In the work of Kerr & Brown (1965), for example, poor weather was encountered during two haymaking experiments. Even with the help of crimping the hay was in the field for 8 to 10 days.

It is the last stages of drying, i.e. from 40 per cent down to about 20 per cent, which are the most difficult to achieve and during which time losses are most liable to occur. In traditional methods of haymaking the 'finishing' of such hay was carried out in loose piles of hay, often built around tripods, which gave some protection from the environment. The hay was allowed to dry out by the effect of wind and a slight natural heating of the stack. In poor weather such methods have been shown to considerably reduce dry matter losses (Murdoch et al., 1959). The cost of producing hay by such labour intensive methods, however, has rendered them unpopular and today hay is normally baled from the swath. Nevertheless,

a similar 'finishing' of hay to that for loose hay may be attempted by leaving the bales in small stacks in the field, but the value of this procedure has not yet been established in experimental work.

A more reliable method of 'finishing' hay is by 'barn' or 'artificial' drying. The bales of hay are built round a duct or over a floor through which cold or slightly warmed air is blown by fan (Culpin, 1962). Hay with initial moisture contents of up to 45 per cent may be safely dried by this method.

Respiration of Stored Hay

Many workers have noted the rapid rise in temperature that occurs in bales of hay soon after they are made (Nelson, 1966; Gregory et al., 1963). Nelson reported that the temperature in bales of Lucerne hay with a moisture content of 48 per cent increased at the rate of 2°C per hour reaching an initial peak of 49°C twelve hours after baling. Hay of 46 per cent moisture was found by Gregory et al., to have reached an initial peak temperature of 48°C in 24 hours falling to 40°C at 48 hours. There is little doubt that these initial peaks are due to the heat produced by the respiring plant cells although, as Gregory points out, certain mould species will already be multiplying.

After cutting, the plant cells of grass continue to live and respire as long as they have a supply of moisture and nutrients. It is only when the moisture content falls below 20 per cent that respiration by the cells finally ceases (Wood and Parker 1971). By the time the hay is baled or stacked the moisture content will have been reduced from the initial figure of 70 to 80 per cent to between 20 and 40 per cent, depending upon the method

and the weather. In most cases therefore, the hay is still actively respiring when it is bulked into storage.

Unless the moisture content becomes limiting the rate of respiration of the plant cells will increase exponentially with the temperature up to 25°C and reach a maximum rate at between 30° and 35°C. The continued heat production by the cells is then likely to raise the temperature further until at about 45°C they die (Wood and Parker, 1971). The initial temperature peak is, therefore, limited by the death of the plant cells rather than by the exhaustion of nutrient reserves.

Some heating has been recorded even in bales at an average moisture content well below the 20 per cent at which plant cells cease to respire (Gregory *et al* 1963). Bales made at 15 per cent moisture heated to 25°C after 48 hours, the temperature then declining to ambient within a few days. However, although at an average moisture content of 15 per cent, it is likely that the bales would contain patches of hay at a higher moisture content which would enable the hay to continue respiring until the moisture content evened out within the bale as a result of the heating.

It is also recognised that handling and bruising of plant tissues, such as that which hay undergoes during baling can cause increases in the rate of respiration which will then fall back to normal within a few days (Stiles and Leach, 1960; Simpson, 1961).

Wood and Parker (1971) found that the plant respiration heat could be used to bring about moisture loss in partly dried bales and that it was in fact a major source of heat in farm barn hay drying systems. It is likely also that the finishing of hay in the field in small stacks of loose or baled hay also depends to some extent on the utilisation of respiration heat to drive off moisture.

Moulding of Stored Hay

At baling the plant material will be contaminated with microorganisms and the spores of fungi of many different types. Even during the first few days of storage, when the main source of heat is from plant respiration, various changes will be taking place in the fungal population. The new conditions will favour the development of a few of the species present and these may develop rapidly to form the 'storage flora', now replacing the old 'field flora'.

Where the moisture content of the hay does not exceed 20 per cent, however, mould growth will be severely restricted although according to Snow et al., (1944 a & b) total inhibition will not take place except at moisture contents in the region of 11 per cent.

In fact the growth of moulds is more closely related to the relative humidity in the immediate environment of the moulds than to the moisture content of the substrate itself (Waite, 1949; Snow et al., 1944a). Observing mould growth on a wide range of feedstuffs over a $3\frac{1}{2}$ year period Snow et al., (1944a) found that the factors controlling mould growth were (1) the relative humidity of the environment; (2) the length of time in storage; (3) the balance and type of nutrients provided by the substrate; (4) the temperature, and (5) the type of mould species present. Mould growth took place relatively quickly on all feeding stuffs stored at 75 to 100 per cent relative humidity. Below 75 per cent relative humidity mould growth, while not prevented, only developed after a very long latent period. Wright (1941) recommended that a relative humidity of 67 per cent should not be exceeded for dried grass storage although at 70 per cent relative humidity 303 days elapsed before visible moulding occurred. Waite (1949) also found

that moulding occurred in cured hay at 70 per cent relative humidity but that at a relative humidity of 65 per cent no moulding occurred after over 400 days. At these low relative humidities moulding was found to be more likely to take place on fodders with a high content of soluble sugars (Snow et al., 1944b).

When left in an atmosphere of constant relative humidity, materials such as hay are found either to gain or loose moisture until they reach a moisture content which is at equilibrium with the atmosphere. Zink (1935) was one of the first workers to establish the relationship between the moisture content of hay and the relative humidity of the environment. Different hays were found to have different equilibrium moisture contents for the same relative humidity and temperature. Dexter (1947) found that immature hays at high relative humidities had higher equilibrium moisture contents than more mature hays, and moulded more quickly than such hays at the same relative humidity. Differences were also observed between fresh dehydrated herbage and dried rehydrated herbage, the former having a higher equilibrium moisture content and being more liable to moulding (Dexter et al., 1947). Dawson and Musgrave (1950) confirmed the above findings but showed differences between herbages to be of greater significance.

The critical moisture content above which most hays will mould is stated by Waite (1949) to be about 15 to 16 per cent at 20°C, this being at equilibrium with a relative humidity of about 75 per cent. However, based on the results of field experiments carried out by Watson in the 1930's, Watson and Nash (1960 d) suggest that a moisture content of 20 to 24 per cent is the most suitable for baling hay. The baled hay did not become visibly mouldy or dusty unless baled at above 32 per cent moisture,

although brown patches due to fermentation did become apparent before this. It would therefore seem possible that even hay which is well preserved by visual assessments may have supported some mould growth. This conclusion is confirmed by the work of Gregory et al (1963) who found growth of mesophilic moulds such as Aspergillus glacus in hays baled at about 25 per cent. Penicillium species were also abundant on hays considered to be 'good' or well preserved. Hay with moisture contents below 20 per cent moisture content were found to have a very sparse microflora even after 3 months storage.

Snow (1949) investigated the ability of fungi isolated from mouldy fodder to grow at a range of relative humidities. Each species was found to have a different range of humidities at which its spores would germinate. The spores of Mucorales and certain pathogenic fungi required high humidities, 93 to 100 per cent. Penicillium had a wider range, from 80 to 100 per cent. Aspergillus species were generally favoured by low relative humidities although some could grow at above 86 per cent relative humidity. The Aspergillus glacus group had the lowest moisture requirement for spore germination and growth, 64 to 70 per cent relative humidity. The ability of this group to exist at low humidities is no doubt responsible for its frequent isolation from a wide range of products stored at sub saturated atmospheres to the exclusion of other moulds which are not so well adapted. Lacey and Lord (1972) noted the presence of Aspergillus glacus on some propionic acid treated hays. Ayerst (1969) has established the temperature and humidity limits for spore germination and growth of a number of fungi commonly found on stored materials.

In batches of hay similar to those used in the First Batch Experiment in this thesis (Section One), made at moisture contents of 38 to 46 per cent, Gregory et al., (1963) found that the growth of mesophilic moulds was rapidly succeeded by thermophilic mould and actinomycetes accompanied by temperatures of 60°C and over. The peak temperature was generally reached in 3 to 7 days but the mould counts continued to rise, due to the reappearance of large numbers of mesophilic moulds, reaching maximums of about 10^7 organisms per gram at 3 to 4 weeks. Characteristic of such hays were the presence of large numbers of the spores of the organisms responsible for Farmer's Lung Disease, Micropolyspora polyspora and Thermoactinomyces vulgaris (Festenstein et al., 1965; Gregory and Lacey, 1963 a). The clouds of spores of these organisms which are released from mouldy bales when they are opened (Gregory and Lacey, 1963 b), give rise to this form of extrinsic allergic alveolitis when they are inhaled into the lungs (Blyth, 1973).

Bales made at 30 to 35 per cent average moisture content were found by Gregory et al., (1963) to be irregularly dried and as a result patches rich in thermophilic actinomycetes existed a few centimeters from much drier parts with Aspergillus glacus moulding. As a result of the heating of baled hay, moisture was lost and irrespective of the initial moisture content, equilibrium was eventually reached at about 15 to 16 per cent moisture. Dry matter losses in the baled hay were attributed mainly to the loss of sugar. It was also noted that the pH of the hay increased where moulding had taken place. When a comparison was made between the course of events in a damp haystack as opposed to baling it was found that the centre of the stack became hotter than the corresponding bales and a silage like bacterial fermentation took place accompanied by a fall in pH.

However, this trend was reversed if moulding developed (Gregory et al., 1963).

Nelson (1966, 1968 and 1972) confirmed the conclusion of Gregory et al., (1963) that moisture content at baling is the most important factor, but found the density of bales also to have a significant effect. Density was found to be an important factor in the bulk storage of chopped hay by Shepperson (1955). Nelson (1972) found that the heat output per pound of dry matter showed a marked increase with increased moisture content but was not affected by increased density at baling. Moulding, however, increased with the density (1966) this being most apparent at 27 per cent moisture when the high density bales moulded while low density bales were free from mould. Loss of dry matter and nutrients, however, were not significantly affected by density (1972).

Losses and Health Hazards Caused by Moulding of Hay

Although dry matter losses of up to 5 or 6 per cent take place in hay baled or stored at moisture contents below 20 per cent, losses are very much increased if the hay is stored at high moisture contents when moulding takes place. Kerr and Brown (1965) found storage losses of 10.8 and 17.5 per cent for hay stored at moisture contents of 31.4 and 39.8 respectively, but only 3.6 for hay stored at a moisture content of 14.9 per cent. Hoffmann and Bradshaw (1937) recorded losses of up to 22 per cent in very high moisture hay. In certain very specific conditions hay baled at high moisture contents can be lost completely as a result of spontaneous combustion (Currie and Festenstein, 1971).

The dry matter loss figures, however, do not indicate the true loss

in value of the hay, which is very much greater as most of the loss takes place from the water-soluble carbohydrate, one of the most digestible portions of the hay (Gregory et al., 1963). Hoffmann and Bradshaw recorded water soluble carbohydrate losses of up to 93.7 per cent in mouldy hay. Other changes associated with the moulding of hay were a rise in the pH value, a loss of lipid soluble dry matter, a rise in the ash content, an increase in soluble nitrogen and a decrease in nitrogen free extractives (Nelson, 1972). Of utmost importance is the fact that where temperatures of above 50°C have taken place in the mouldy hay its digestibility is likely to fall, due to the denaturing of the protein (Watson and Nash, 1960 e).

Although the nutrient and digestibility losses caused by moulding are of considerable economic importance to the farmer, current interest has also centred on the occurrence of Farmer's Lung Disease antigens in mouldy hay (Gregory et al., 1964). In a study of the farming population in parts of Scotland the incidence of Farmer's Lung Disease in the North and West was found to be remarkably high with 86 per 1000 of the farming population affected (Grant et al., 1972). Of these, 54 per cent were caused through the handling of mouldy hay.

According to Lacey (1964), when a bale of mouldy hay is opened the concentration of the spores of Micropolyspora polyspora and Thermoactinomyces vulgaris, which are responsible for the allergic condition, may reach over 1500 million per cubic metre. Gregory et al., (1963) reported that hay baled at moisture contents over 35 per cent are likely to create the greatest risk of causing Farmer's Lung Disease. Conditions similar to Farmer's Lung Disease have also been reported in dairy cattle (Pirie et al., 1971) and in sheep fed on mouldy hay (Lacey et al., 1972).

It is also believed that the feeding of mouldy fodder can give rise to the form of mycosis causing mycotic abortions (Ainsworth and Austwick, 1955 a & b).

Certain moulds liable to grow on mouldy hay are able to produce toxins which can have serious effects on animals eating the hay. One such mould is Aspergillus fumigatus (Lynch, 1972). In Russia (Ursiny et al., 1964) the death of 268 cattle was reported to be due to the feeding of mouldy hay and other fodders containing mycotoxins. Symptoms such as diarrhoea, fever, dullness, inappetite and abortion were reported in this outbreak. It is more common, however, for mouldy hay to result in poor performance of the animals feeding on it, without the toxins being specifically implicated. In the experiments of Mohanity (1968) in the USA, the feeding of mouldy fodder resulted in lower dry matter intakes and reduced weight gains. In England, Burt et al., (1964) traced the poor milk yield of a group of dairy cattle to the feeding of fodder, which, by chemical analysis was satisfactory, but which carried a high level of mould contamination and proved to be of low digestibility in feeding trials. It is clear, therefore, that even where fodder is not of low nutritional status due to moulding, it may still present a serious health hazard both to the farm workers handling it and to the animals receiving it.

SECTION ONE

FIRST BATCH EXPERIMENT

AIM

The aim of this experiment was to establish the levels of propionic acid required to prevent moulding and to preserve hay baled at different moisture contents, as far as possible under commercial farming conditions. The effects of the various treatments were observed on batches of bales stored in a large, well ventilated shed. Records were kept of the changes in temperature, moisture content, water soluble carbohydrate, pH and mould counts. Dry matter and water soluble carbohydrate losses were calculated.

A randomised block design was chosen comprising three moisture levels and five acid treatment levels, including an untreated control. Two replicates were intended for each treatment. It was decided that moisture levels of 25, 35 and 45 per cent should be aimed at, with propionic acid treatments of nil, 1, 2, 3 and 4 per cent by weight to be applied to the bales at each moisture level. In view of the difficulty of measuring the moisture content of hay in the field, and the impossibility of measuring the moisture content of the swath intended for baling at any one instant, it was not known how accurately these moisture levels could be achieved. Similarly, as the accuracy of the acid applicator was not known, the acid treatment levels had to be approximate aims rather than absolute values.

The hay was to be made in the field and sprayed with the propionic acid using a prototype applicator designed and supplied by BP Chemicals (International) Ltd., and fitted to the baler. Sufficient bales were to be made to provide 9 bales in each of the two replicate

batches of each treatment. At the lowest moisture level a further two replicate batches of each treatment were to be made and left in the field for a few weeks before being brought into storage. All the other batches were to be brought into storage immediately after baling, and the monitoring of changes begun.

It was hoped that the batches left in the field would provide an interesting comparison in view of the common farming practice of leaving such small stacks in the field after baling, presumably to promote further drying of the hay.

METHODS

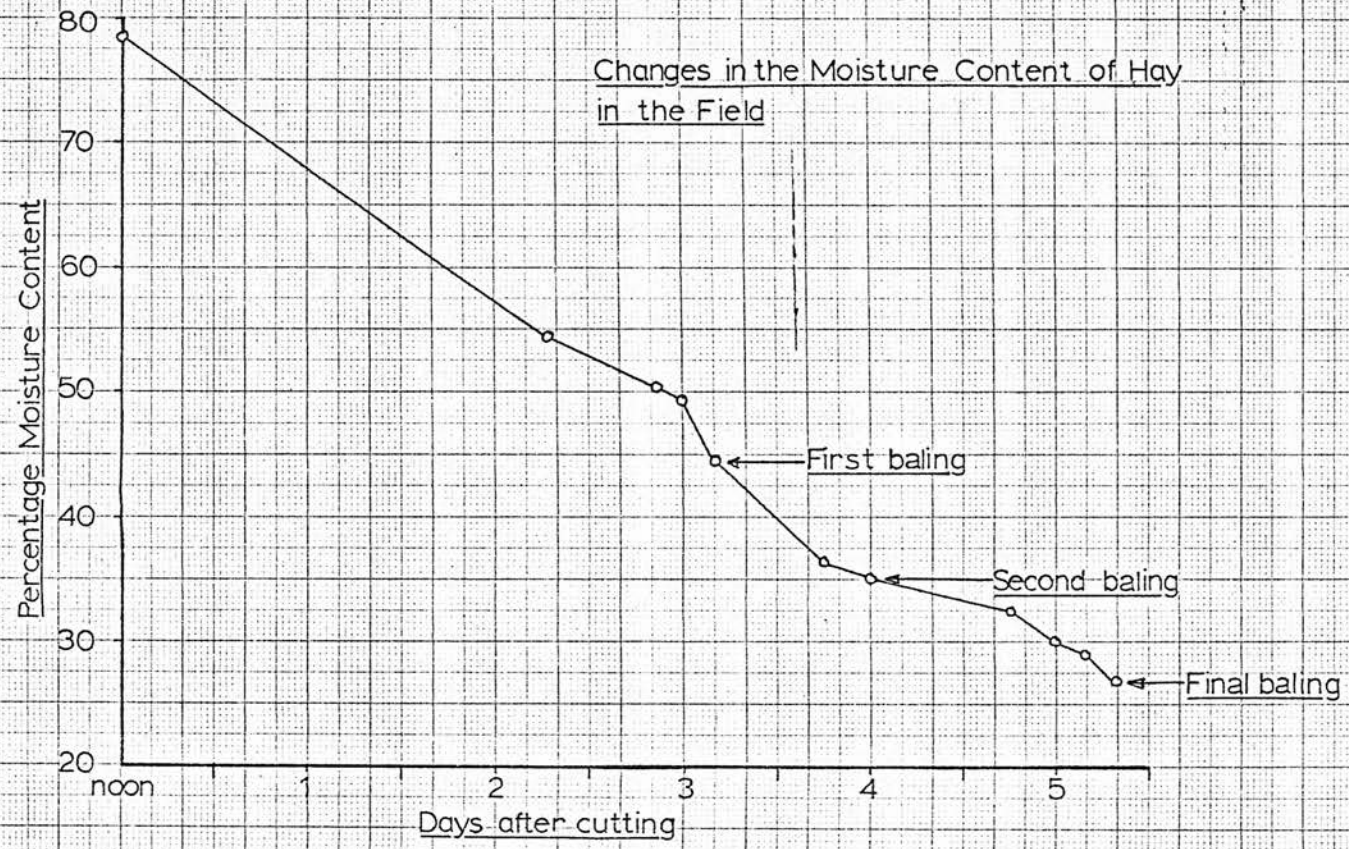
Field and Storage Work

An initial attempt to carry out the experiment was begun on June 29th 1970, but had to be abandoned when the acid applicator broke down. After repairs had been carried out another field was made available and a fresh start was therefore made on July 13th.

Hay was made from 8 acres of a 2-year ley largely comprising ryegrass and red clover. The crop was cut using a reciprocating mower. Tedding of the hay commenced on the day after cutting, and tedding and turning were carried out in accordance with the established techniques of fast haymaking, but without crimping. Immediately prior to baling, the hay was windrowed by running two swaths into one which was then of a suitable size for baling.

As the moisture content of the hay fell the moisture values were plotted on a graph against time (Figure 1). Thus, providing the weather conditions did not vary greatly it could be predicted when the

FIGURE 1


FIRST BATCH EXPERIMENT
Changes in the Moisture Content of Hay in the Field
Percentage Moisture Content
80
70
60
50
40
30
20
noon
1
2
3
4
5
Days after cutting
First baling
Second baling
Final baling

hay would dry to the desired moisture levels. Good weather conditions prevailed during the 2nd and 3rd days after cutting, and baling commenced at 4pm on the third day, by which time the moisture content had fallen to 44 per cent.

The first 9 bales to be used for the experiment were not treated with propionic acid and formed the first control batch. Eighteen bales treated with approximately 4 per cent propionic acid were then made and the bales randomly allocated to the two replicate batches of 9 bales. This process was repeated for the 3 per cent, 2 per cent and 1 per cent treatments in that order. As the 1 per cent treated bales were being made rain began to fall which prevented the baling of a further 9 untreated bales. These bales were to have formed the 2nd replicate control batch. When baling was completed the bales were carted to the shed, individually weighed using a spring balance, and built into identical batches of the design shown in Figure 2. The arrangement of the batches in the shed is given in Figure 3.

Drying weather conditions continued during the 4th and 5th days after cutting, and the baling of the 35 per cent moisture and 26 per cent moisture bales was carried out on the afternoons of these days in succession. The baling was carried out in the same way as described for the highest moisture level, and bales made each day were built into their corresponding batches that evening.

At the 26 per cent moisture level twice as many bales were made for each treatment, the bales for immediate storage being selected randomly and the others built into similar batches in the field. Plastic bags were used to cover the top of these batches to prevent penetration of rain. After a four week period the field batches

FIGURE 2 Design of the 9-bale Batches

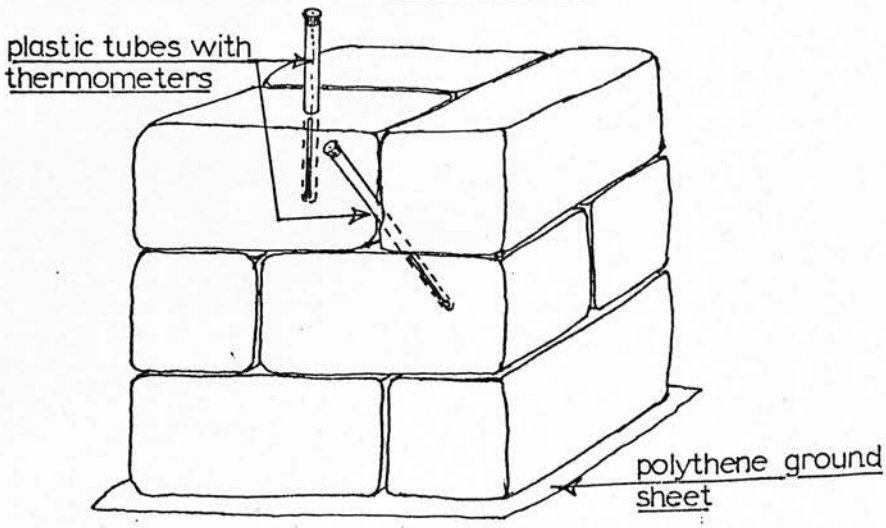
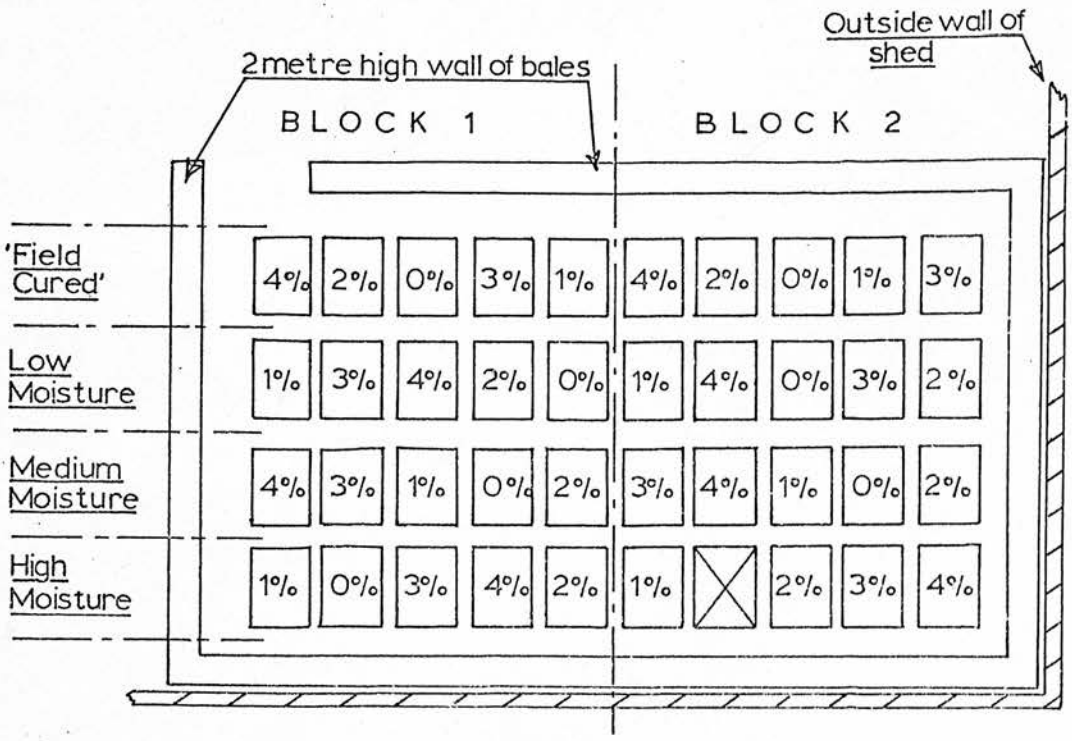


FIGURE 3 Arrangement of Batches in First Batch Experiment



were taken into the shed and kept with the other batches until the end of the experiment.

In the storage shed which was large and airy an area of about 20 metres by 15 metres was allocated to the experimental batches. Ideally the batches should have been arranged in two 'blocks', each containing one batch from each of the treatment levels, with all the batches in each block arranged randomly. In practice, however, a completely random arrangement was not possible due to the difficulty of lifting bales over batches which had already been built. If all three moisture levels had been baled on the same day no problem would have arisen, but the bales at each moisture level were made and built into the batches on different days. Randomisation therefore could not be carried out between the batches at the different moisture levels, but only between batches at the different acid levels in each moisture level. Figure 3 shows that the bales were arranged in two blocks each with 4 rows of bales, a row for each of the three moisture levels, and a row for the bales brought into storage after being left in the field for a few weeks.

This arrangement of the bales limited the statistical analysis in that the true effect of moisture content could not be separated from the effect of the position of the bales in the shed. Levels of significance could therefore only be obtained for the differences between acid treatments and not for the differences between moisture contents. It was hoped that the effect of the rows being in different positions on the final outcome would be minimal, so that it could be safely assumed that the final differences between the batches in the rows would be principally due to the effect of the moisture content.

Experimental Methods

Sampling

The crop was sampled at the time of cutting for In vitro digestibility of the organic matter and for water soluble carbohydrate content. During the drying period regular samples were taken for moisture determinations. For all these samplings the standard method of sampling was used in which 20 to 25 grab samples were taken from the swaths which were crossed while the person taking the samples walked in a 'W' path over the field. Before taking grab samples, the section of the swath to be sampled was thoroughly mixed to eliminate variation between the top and bottom of the swath.

At the time of baling a number of grab samples, giving about 1 kg of hay, were taken from the swath immediately in front of the baler. This was carried out separately for each treatment in the experiment and the samples were analysed the same day for moisture content and water soluble carbohydrate. In addition one bale which had received an acid treatment was set aside and subsequently sampled in order to study the distribution of acid within the bale.

During storage, sampling of each batch was carried out at two weeks and six weeks and at the termination of storage, ie. after 12 to 14 weeks. These samples were obtained using a specially made bale corer consisting of a 30cm long steel tube of 2.5cm internal diameter with a stainless steel cutting edge at the tip which was sharpened before taking cores. The corer was powered by a 3/4 inch electric drill and based on the designs of Simmons (1961) and Bratzler and Washco (1959) and is shown in Figure 4.

For the 2 week and 6 week samplings, cores were taken from the

FIGURE 4

Corer used during the First Batch Experiment

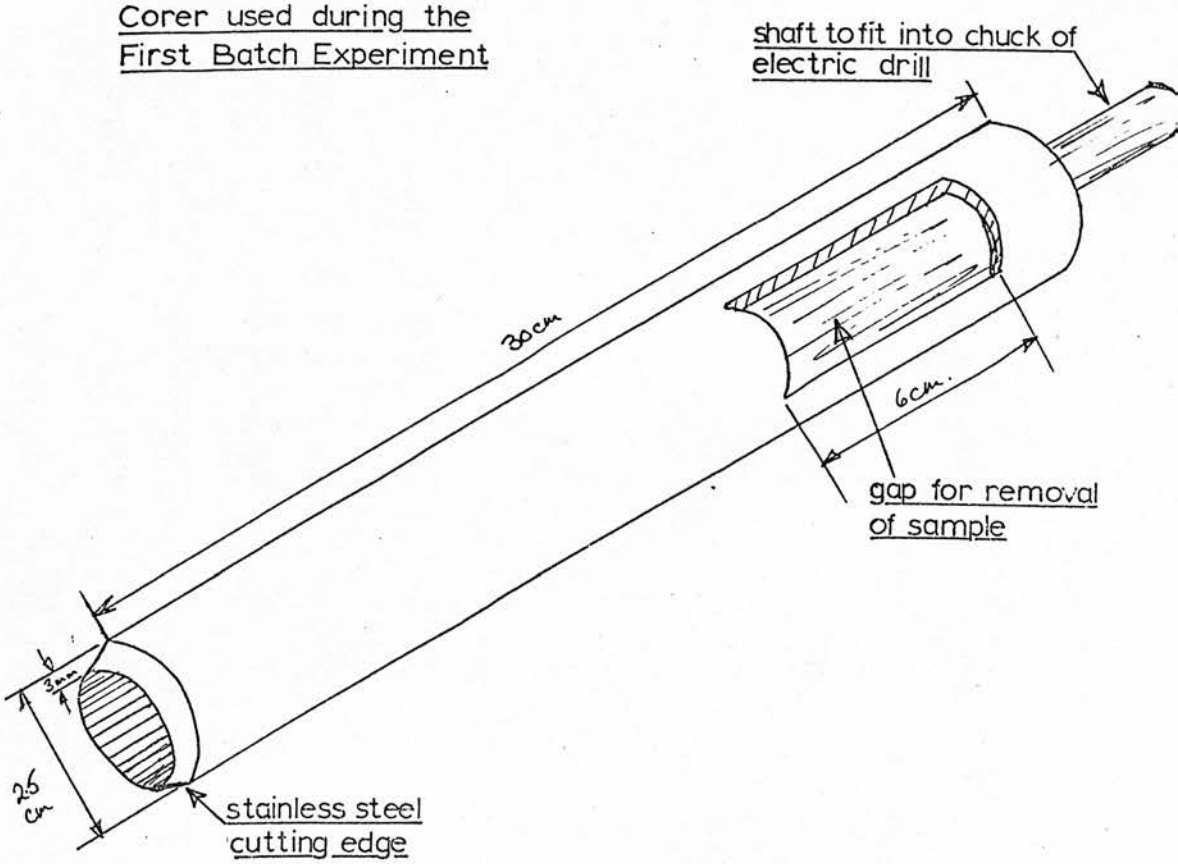


FIGURE 5 BALE, SHOWING THE METHOD OF SAMPLING

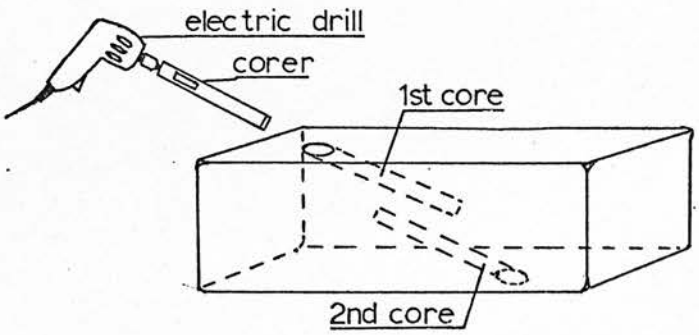
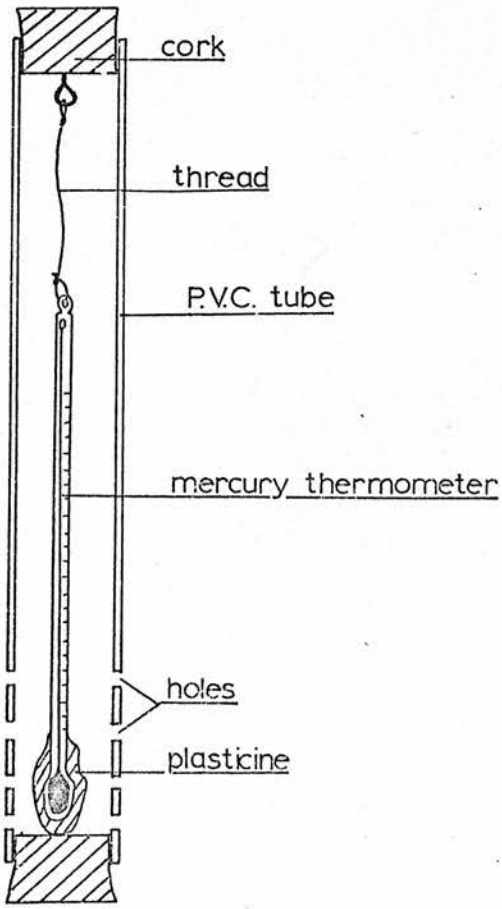


FIGURE 6

THERMOMETER IN TUBE AS USED IN BATCH EXPERIMENTS



same position in each batch, two cores being taken from each of two bales in the batch (Figure 5). The batches were disturbed as little as possible during the operation, to prevent the temperatures in the batches being affected. The cores from each batch were then mixed and subdivided into two samples, one for microbiological and one for chemical analysis.

The final sampling involved taking two cores from every bale and was carried out over a three week period, one week being needed for each moisture level. The bales in each batch were first weighed using a spring balance, and then cored, two cores being taken from each bale. The sample from each bale was mixed and divided into two, part going towards a bulked sample for the batch, and part towards a moisture determination for the bale.

Temperature Readings

Temperature readings were recorded from two locations in each batch (see Figure 2) throughout the storage period. As each batch was being built plastic tubes of about 2cm diameter were inserted into the corresponding two bales in each batch so that the tip of the tubes were at about the centre of the bales. When the batch was complete, mercury in glass thermometers was inserted down the tubes (Figure 6). A piece of plasticine was placed over the bulb of each thermometer to insulate it slightly and prevent inaccurate readings when the thermometer was pulled out of the tube. The thermometers could be easily withdrawn by means of a thread attached to the cork sealing the end of the tube. Readings were taken twice daily during the first week to ten days of storage but the interval between readings then increased as the fluctuations in temperature became more gradual.

In addition to the thermometers already mentioned, an extra supply of thermometers made it possible to put a thermometer into each bale of five of the batches. This was carried out in one control (untreated) batch at each moisture level, in a medium moisture batch which had received 2 per cent acid and in a high moisture batch which had received 1 per cent acid. About 120 thermometers were used in the experiment altogether.

Temperature readings in the field cured bales were not taken until after the bales had been brought into storage.

Propionic acid Application

The acid was applied using a prototype applicator supplied by BP Chemicals (International) Ltd., and fitted to an International B47 baler by the East of Scotland College of Agriculture Farms Division Workshop. A full description of the applicator is given in Appendix 7. Before being used the calibration of the applicator was checked against the graph supplied with the applicator and shown in Appendix 7.

Before baling at each of the acid treatment levels the applicator had to be set to provide as accurately as possible the desired acid level. This was done using the following procedure:-

a. The rate of baling was estimated by timing the production of 5 or 6 bales, weighing these bales, and calculating the rate in kilograms of hay per minute.

b. From the rate of baling the rate of flow of acid for the appropriate acid level, 1, 2, 3 or 4 per cent was calculated.

PLATE 1.

The BP Chemicals high pressure applicator in use

PLATE 2.

The high pressure applicator with cover removed

PLATE 3.

The baler and high pressure applicator before the first season's work

PLATE 4.

The same baler after one season's work showing the paint stripping caused by the acid

c. Using the calibration graph shown in Appendix 7 the applicator was set to provide the required rate of acid.

By using different tractor speeds together with the range of adjustment on the applicator itself it was possible to encompass all the acid levels required except the highest acid level on the high moisture hay. This treatment fell short of the 4 per cent level as the applicator could not supply acid at a fast enough rate.

The exact quantity of acid used at each treatment level was recorded and used together with the weights of the treated bales to calculate the actual average treatment levels.

Moisture Content Determinations

Moisture determinations were normally carried out using the standard oven technique of drying the samples for 24 hours in an oven at 100°C. Where small samples were being used the cooling of samples before the final weighing was carried out in a desiccator. All analyses were carried out in duplicate unless otherwise stated.

On a number of occasions, particularly when the decision to start baling was involved, a quick result rather than a very exact one was important. For these determinations a specially modified hot air fan drier was used in which 20 to 30 grams of chopped sample was placed in a mesh bottom tray through which a gentle current of warm air was blown by means of a fan. The results were obtained in a half to one hour and were within 3 per cent of the oven dry matter result. A full description of the hot air fan drier is given in Appendix 8.

Chemical Analyses

Chemical analyses were commenced on the day of sampling. Dry matter determinations were made on 2g to 5g samples dried for 24 hours at 100°C and cooled for 10 minutes in a desiccator before weighing. Water soluble carbohydrates were determined following the method of McDonald and Henderson (1964), a full description of which is given in Appendix 10.

The pH values of the hay samples were taken using a 'Pye universal' pH meter after a 10g sample had been macerated with 250ml of water. All analyses were carried out in duplicate.

Visible Mould Assessment

While the analysis of cored samples may give a reasonably accurate picture of the average condition of the bales in terms of moisture content, water soluble carbohydrate and pH, which is their purpose, this particular investigation required further information. The above factors are merely the symptoms, from which the presence of causal organisms can be deduced, and the extent of the damage estimated.

Only a visual assessment of the extent and pattern of moulding within the bales would give the additional information such as:-

a. How widespread the moulding is within each bale

b. The nature of the moulding occurring at different moisture levels and with different acid treatments.

This information could be valuable in establishing how efficiently the applicator was distributing the acid within the bales

and to what extent the moisture levels and acid treatments were influencing the pattern of moulding in the bales.

The cored samples taken at the first sampling date were used for microbiological analysis but this could only provide an average for each bale. Furthermore, these results, for reasons to be discussed later, are not particularly reliable when it comes to the fungal counts and are unlikely to give a true picture of the relative moulding in different bales.

For the visual assessment two bales were chosen randomly from each batch at the end of the storage period. Each of these bales was opened in turn and divided into 8 or 9 sections and from a visual examination of these the proportion of mouldy hay in the bale as a whole was estimated. Based on this figure the bale was given a score according to the following scale:-

0 - No visible moulding

1 - Under 25 per cent of the bale with visible moulding

2 - Between 25 and 50 per cent of the bale with visible moulding

3 - Over 50 per cent, but less than 100 per cent of the bale with visible moulding

4 - Mould throughout the bale

5 - Extreme moulding and discolouration of the bale.

In the low moisture bales, moulding, when present, gave a dry dusty appearance to the bales but did not result in clearly defined patches of mould. Such bales did not easily fit into the grades given above and so were classified as being 'dusty' or 'not dusty'.

In order to give some evaluation of the different types of

moulding which had taken place the bales at each moisture level were further examined and samples were taken for detailed microbiological examination. These samples were not taken randomly, but were chosen carefully to represent, as far as possible, the different degrees and types of mould contamination seen by visual examination. Each sample consisted of about 500g of hay removed by hand from the bales. The samples were put through the microbiological tests as outlined below.

Microbiological Analyses

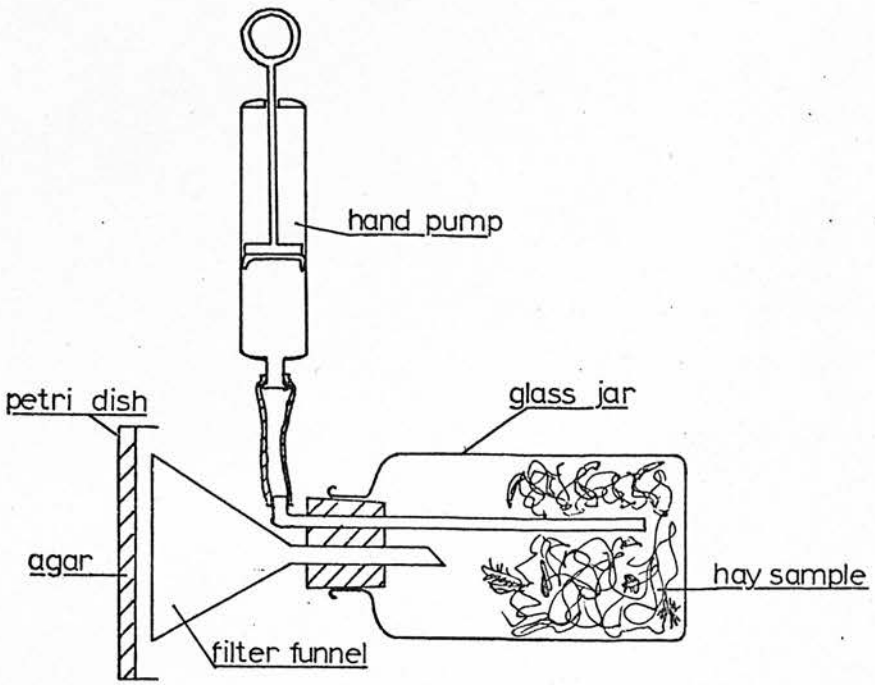
The cored samples taken at the first sampling date were analysed using the serial dilution technique which is outlined in Appendix 12. Mould counts were made on malt salt agar incubated at 28°, 37° and 55°C. Counts were taken at 3 and 5 days.

This method was found, however, to take up a disproportionate amount of time and was not used on the cored samples taken at the second sampling date or at the end of storage.

For the selected samples taken from the bales after the visual examination at the end of storage a method was devised which, it was hoped, would give a reasonably reliable comparison between the samples in terms of mould contamination although not giving such a good picture of the numbers of organisms present. Following the example of Gregory and Lacey (1963)a, but without the facilities to use a wind tunnel and Anderson sampler, a simpler device was made and tested which by the use of a small hand pump blew spores off a sample of hay in a jar onto an agar plate (Figure 7).

Five to 10 grams of the hay sample were placed in the jar which was shaken for 20 seconds by a mechanical shaker. The hand pump was

FIGURE 7 AIR DISPLACEMENT SAMPLER



then used to displace a fixed volume of air from the jar through an inverted filter funnel over the end of which was held a petri dish containing a suitable media. Spores released from the sample by the shaking were carried out in the air current and impacted onto the surface of the media. The petri dish was immediately covered and incubated at the desired temperature. Where more than two plates were being made from one sample, the jar was shaken between each two plates.

The results of tests with this sampler are given in Appendix 11.

RESULTS

The three moisture levels at which the hay was baled were near to the intended levels, as will be seen from Table 1. The levels of propionic acid treatment were, however, on the whole slightly high. An exception to this was the 4 per cent acid level at the highest moisture level which could not be achieved due to the limitations of the applicator.

TABLE 1

Moisture Content			Acid Treatment	
Intended	Achieved (Bulked)	Individual Batch	Intended	Achieved
45%	43.4%	41.6%	4%	3.5%
		41.1%	3%	3.2%
		45.5%	2%	2.1%
		44.2%	1%	1.1%
		44.5%	Control	-
35%	35.6%	39.6%	4%	4.1%
		33.5%	3%	3.3%
		34.3%	2%	2.3%
		36.0%	1%	1.0%
		34.6%	Control	-
25 - 30%	26.7%	26.4%	4%	4.5%
		27.5%	3%	3.4%
		26.8%	2%	2.4%
		26.5%	1%	1.0%
		26.5%	Control	-

MAIN EFFECTS

The results are presented so that the main variable factors, namely the initial moisture content (3 levels) and the propionic acid treatment rate (5 levels), are considered together as to their effect on the initial bale weight, on the digestibility, the moisture content and the water soluble carbohydrate content of the hay at the end of the storage period, and on the losses of moisture, dry matter and water soluble carbohydrate during the storage period.

The effects of initial moisture content and of the acid treatments on the pattern of changes during storage are then presented in the light of the temperature readings and the results of the samplings carried out at 2 weeks and 6 weeks after the start of storage.

a. On the Initial Bale Weights

TABLE 2

Mean Initial Bale Weights (Kg)

	NIL	1% acid	2% acid	3% acid	4% acid	Mean
43.4% moisture	38.2	34.1	33.7	33.7	33.8	34.3
35.6% moisture	33.4	30.4	28.4	28.8	29.4	30.1
26.7% moisture	27.1	26.5	25.0	25.2	26.7	26.1
Mean	31.8	30.3	29.0	29.3	29.9	30.0

Least significance difference between acid treatment means 1.2 kg. ($P = 0.05$)

All the acid treatments resulted in bales which were significantly lighter than the untreated bales, but there was little variation in

weight between the bales at the different acid levels. The weight difference between treated and untreated bales was most marked at the medium and high moisture levels.

It can also be seen from the table that there was a clear trend for the bale weight to increase as the moisture level became higher.

b. On the Percentage Moisture Content at the end of storage

TABLE 3

Percentage Moisture Content

Initial	Nil	1% acid	2% acid	3% acid	4% acid	Mean
43.4% moisture	19.70	23.53	28.30	28.35	30.01	26.75
35.6% moisture	17.34	20.66	25.83	30.56	30.10	24.90
26.7% moisture	15.73	22.98	27.43	27.13	29.11	24.47
Mean	17.31	22.39	27.18	28.67	29.74	25.33
*Field Cured	19.05	20.06	23.65	23.55	25.20	22.41

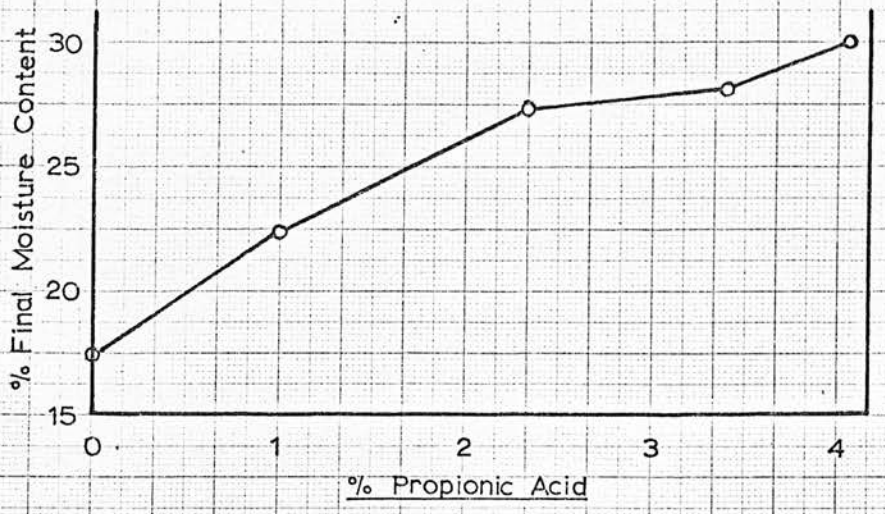
Least significant difference between acid treatment means 2.98% (P = 0.05).

*Not included in the statistical analysis.

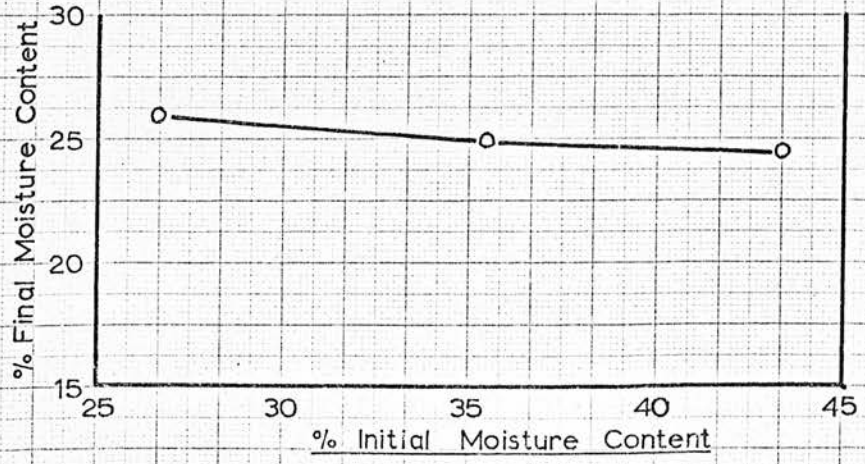
By the end of the three month storage period the moisture content of the untreated bales at all three moisture levels, and of the treated bales at all four acid levels on the medium and high moisture hays, had fallen significantly below the initial moisture content. The moisture contents of all the untreated batches had fallen to below 20 per cent by the end of storage, with only a 4 per cent difference between them.

FIGURE 8 MOISTURE CONTENT AT END OF STORAGE

A Effect of Propionic Acid on Mean Final Moisture Content



B Effect of Initial Moisture Content on Final Moisture Content



At the start of the experiment there had been a difference of over 15 per cent. Looking at the mean effect of the initial moisture content, it is clear that the initial level seemed to have little influence on the final moisture content of the hay (Figure 8B).

The various acid treatments had more influence on the outcome however (Figure 8A), as the 1 per cent and 2 per cent acid levels resulted in moisture contents significantly higher than in the contents at the end of storage. The 3 and 4 per cent acid levels brought little or no further change however. It should be noted that in the low moisture hay the moisture contents at the 2, 3 and 4 per cent levels did not change significantly during storage, whereas at the medium moisture level a drop of at least 5 per cent, and at the high moisture level, of at least 10 per cent, took place at all these treatment rates.

The results of the field cured bales, although not included in the statistical analysis, show that a similar trend occurred in these bales to that which occurred in the low moisture stored bales. The field cured bales had a lower final moisture content than the stored bales, except in the untreated batches, with the result that the difference between the treated and untreated bales was much less marked in the field cured bales.

C. On the Loss of Moisture

TABLE 4

Percentage Loss of Moisture

	Nil	1% acid	2% acid	3% acid	4% acid	Mean
43.4% moisture	72.90	67.28	58.06	58.22	60.92	62.42
35.6% moisture	65.71	58.21	40.41	25.54	36.29	45.35
26.7% moisture	53.69	31.68	4.20	13.85	5.33	21.75
Mean	62.34	52.39	34.22	32.54	34.39	42.51

Least significant difference between acid treatment means 10.63% (P = 0.05)

The moisture loss figures presented in table 4 reflect the moisture content changes which have been discussed in the previous paragraph. Over the whole range of acid treatments the high moisture hays lost more moisture than the medium moisture hays which in turn lost more than the low moisture hays. The very high losses recorded in all the untreated batches and in most of the medium and high moisture treated batches should be considered in the light of the high temperatures recorded in these batches (see Figures 11 and 12).

Following the same pattern as the moisture content changes, the 1 and 2 per cent acid treatments progressively reduced the loss of moisture at all three moisture levels (95% significance), this trend being most marked at the lowest moisture level. In all the hays the moisture losses from the 3 and 4 per cent treatments were not significantly different from those of the 2 per cent treatment level.

d. On the Percentage Water Soluble Carbohydrate at the End of Storage

TABLE 5

Percentage Water Soluble Carbohydrate in Dry Matter

	Initial	Final				
		Nil	1% acid	2% acid	3% acid	4% acid	Mean
43.4% moisture	16.5	2.73	3.16	4.89	4.54	5.70	4.37
35.6% moisture	15.1	3.70	6.29	7.08	11.60	10.23	7.78
26.7% moisture	14.5	6.51	10.23	14.32	13.51	15.57	12.03
Mean	15.4	4.63	6.57	8.76	9.88	10.50	8.19
*Field Cured	14.5	5.44	7.32	10.21	9.05	9.41	8.29

Least significant difference between acid treatment means 2.16% (P = 0.05)

*Not included in the statistical analysis.

A comparison between the initial water soluble carbohydrate values and those at the end of storage reveals that in most cases a decline in the percentage water soluble carbohydrate took place. These changes will also reflect, however, the changes in other components of the dry matter and so it is better to look at the effects of the treatments on the water soluble carbohydrate losses (Table 6) than to look at the changes in water soluble carbohydrate content. For the field cured bales loss figures could not be calculated as the initial bale weights were not taken in the field, but from the results of the water soluble carbohydrate determinations included in table 5 and in Figure 9A it will be seen that these bales had a mean water soluble carbohydrate level almost 4 per cent lower than in the corresponding bales which had been brought into storage immediately after baling. This trend is present in the untreated bales and at all the acid treatment levels.

e. On the Water Soluble Carbohydrate Losses

TABLE 6

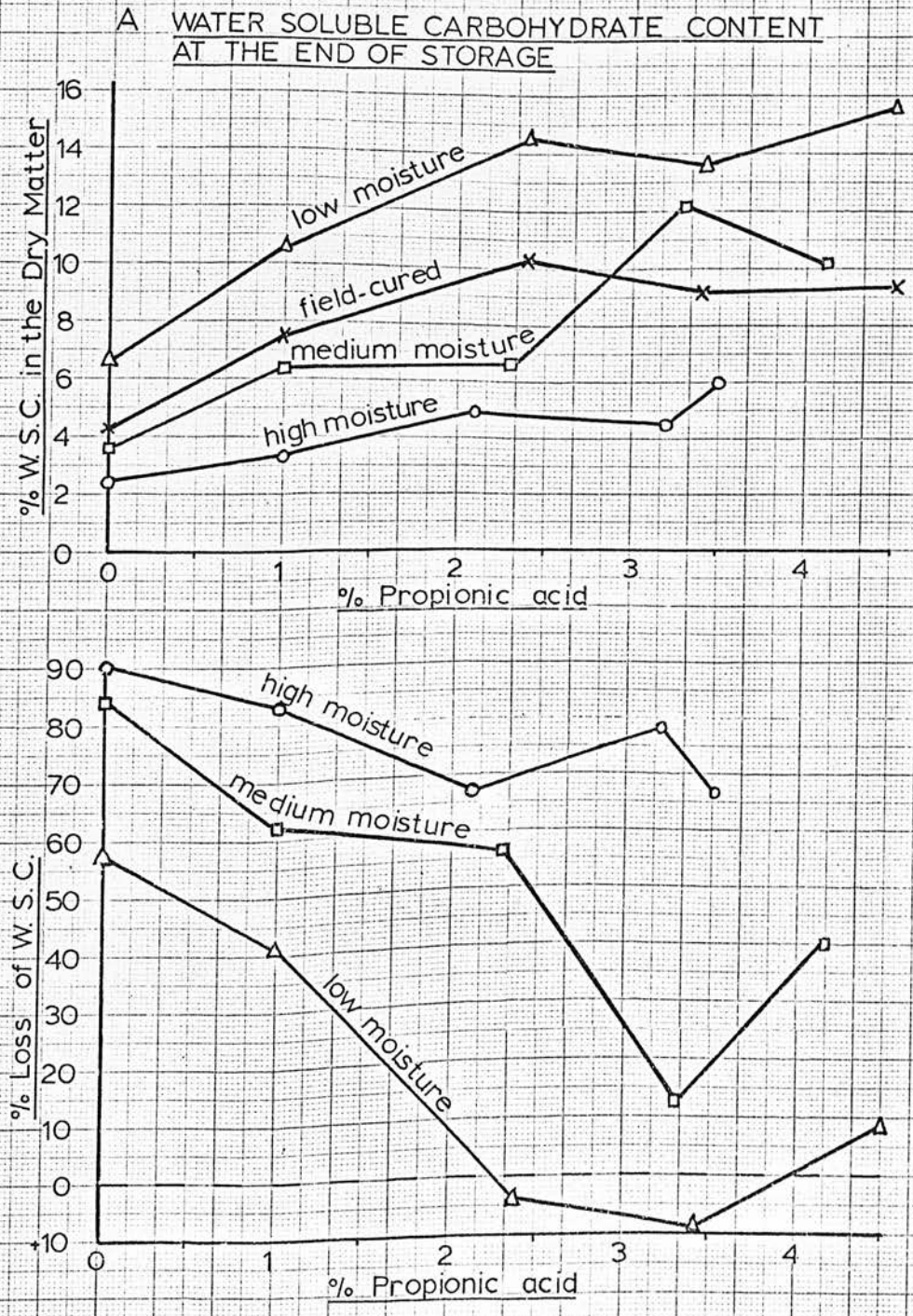
Percentage Water Soluble Carbohydrate Losses

	Nil	1% acid	2% acid	3% acid	4% acid	Mean
43.4% moisture	88.32	82.77	66.76	78.37	66.31	75.14
35.6% moisture	83.61	62.50	56.46	12.74	40.10	51.08
26.7% moisture	56.92	40.82	-3.07	-8.98	8.32	18.80
Mean	73.88	61.95	40.05	27.38	38.24	47.42

Least significant difference between acid treatment means 13.3% (P = 0.05).

At all three moisture levels the loss of water soluble carbohydrate in the untreated bales was high, ranging from almost 57 per cent

FIGURE 9


A WATER SOLUBLE CARBOHYDRATE CONTENT AT THE END OF STORAGE
% W.S.C. in the Dry Matter
16
14
12
10
8
6
4
2
0
low moisture
field-cured
medium moisture
high moisture
0
1
2
3
4
% Propionic acid
% Loss of W.S.C.
90
80
70
60
50
40
30
20
10
0
+10
high moisture
medium moisture
low moisture
0
1
2
3
4
% Propionic acid
B WATER SOLUBLE CARBOHYDRATE LOSSES DURING STORAGE

in the low moisture hay to almost 90 per cent in the high moisture hay. The 1 and 2 per cent acid treatments at each moisture level reduced the losses progressively (significant at 95%), cutting them to nil at the lowest moisture level and by 26 per cent and 22 per cent at the medium and high moisture levels respectively.

The 3 per cent acid level brought a further improvement in the conservation of water soluble carbohydrate only in the medium moisture hay while in the low moisture hay a slight gain of water soluble carbohydrate was recorded and in the high moisture hay the water soluble carbohydrate loss increased. The 4 per cent treatment level brought increased losses in the medium and low moisture hays, but the 3.5 per cent acid treatment reduced the losses in the high moisture hay to 61 per cent of the original water soluble carbohydrate (Figure 9B).

f. On the Loss of Dry Matter

TABLE 7

Percentage Dry Matter Losses

	Nil	1% acid	2% acid	3% acid	4% acid	Mean
43.4% moisture	17.56	14.47	10.42	7.80	8.89	11.19
35.6% moisture	13.94	8.78	12.41	8.73	2.61	9.29
26.7% moisture	7.43	7.40	4.65	1.04	3.61	4.82
Mean	12.06	10.21	9.16	5.85	5.03	8.34

Least significant difference between acid treatment means 3.71% (P = 0.05)

At all three moisture levels the 3 and 4 per cent acid treatments resulted in dry matter losses which were significantly below those in the untreated control bales. The 1 and 2 per cent acid treatments did

not lead to reduction of the losses to a statistically significant extent in most cases, but showed the same trend for losses to be lower as the acid treatment became higher.

In the untreated bales and at all the acid treatment levels the dry matter losses rose as the moisture content increased. At the highest moisture level the 3 per cent and 4 per cent treatments reduced the losses below 10 per cent, while in the medium moisture bales the 4 per cent treatment brought the dry matter loss as low as 2.6 per cent. It was only in the low moisture hay treated with 3 per cent acid that the dry matter loss fell to a value not significantly above zero.

The mean results from the above table are along with those of the water soluble carbohydrate mean losses presented in Figure 10.

g. On the In vitro Digestibility at the End of Storage

TABLE 8

In vitro Digestibility Percentage

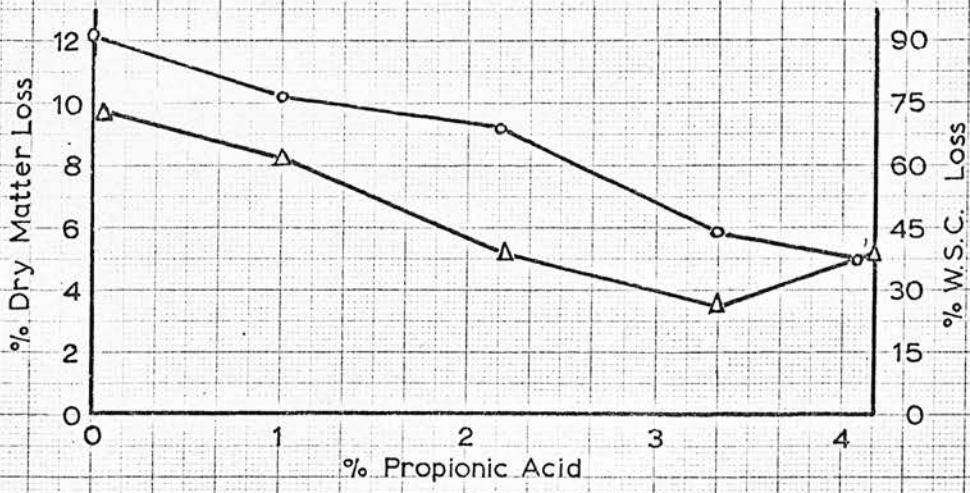
	Nil	1% acid	2% acid	3% acid	4% acid	Mean
43.4% moisture	56.5	58.6	59.8	57.6	59.8	58.6
35.6% moisture	58.7	62.6	65.6	62.1	61.6	62.1
26.7% moisture	60.8	63.6	64.5	65.0	66.4	64.0
Mean	59.1	61.6	63.3	61.5	62.6	61.5
Field Cured	58.9	58.8	61.1	60.8	61.2	60.2

Mean Initial Digestibility 63.0%

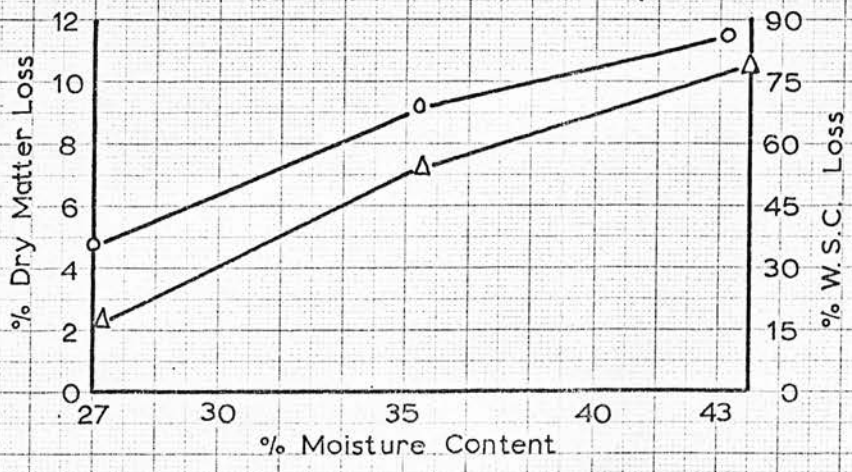
Least significant difference between acid treatment means 3.5% (P = 0.05)

FIGURE 10 PERCENTAGE LOSSES OF DRY MATTER AND WATER SOLUBLE CARBOHYDRATE AT END OF STORAGE

A EFFECT OF PROPIONIC ACID ON MEAN LOSSES



B EFFECT OF MOISTURE LEVEL ON MEAN LOSSES



Dry Matter Losses o W.S.C. Losses Δ

Only at the 2 per cent acid level did the acid treatment result in a digestibility significantly above that of the untreated hay at the end of storage. However looking at the different moisture levels, it was only at the medium moisture level that this result was significant, and even at this moisture level the higher acid levels did not result in any significant difference in digestibility. When the mean results at each moisture level are compared a tendency for the digestibility to fall with increasing moisture level can be seen. The digestibility of the field cured hays was below that of the corresponding stored bales at each acid treatment level.

PATTERN OF TEMPERATURE CHANGES

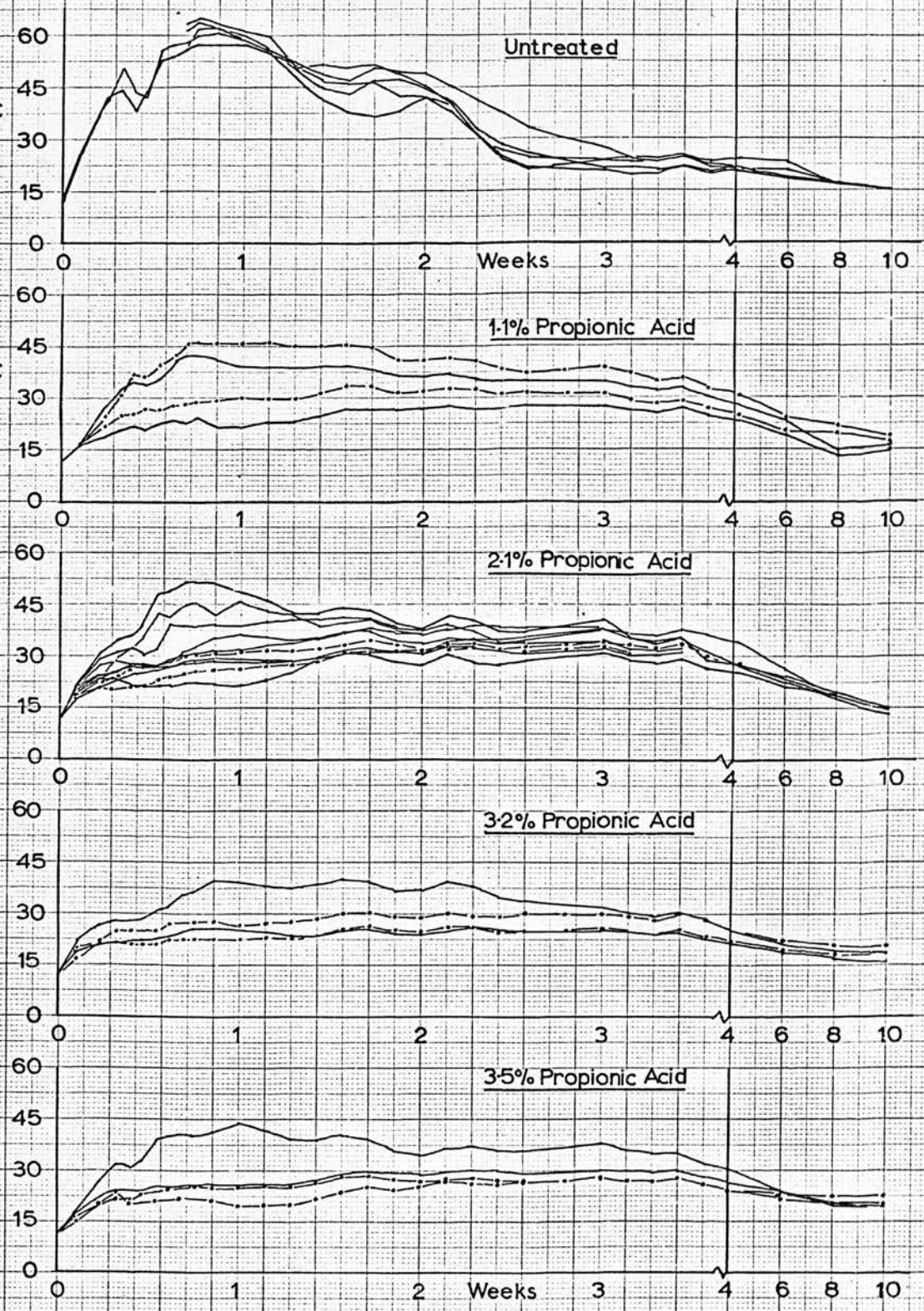
a. High Moisture Hays

The effect of the propionic acid treatments on the heating of the bales may be seen by comparing Figures 11, 12, 13 and 14. At the highest moisture level (Figure 11), rapid heating took place in the untreated bales, an initial peak being reached on the 3rd day.

Between the 3rd and the 4th days the temperature fell by a few degrees and then rose to a secondary peak during the 5th and 6th days. It was only in these untreated high moisture bales that temperatures of over 60° celcius were reached. From the end of the first week onwards the temperature declined to reach the ambient during the third week. During day 5 three additional thermometers were located in the batch in different bales. The temperature pattern in each of the five bales being monitored was similar.

The high moisture hays which had received acid treatments did

FIGURE 11 HIGH MOISTURE HAY



Each line represents one thermometer.
Both replicate batches included in every graph.

FIGURE 12 MEDIUM MOISTURE HAY

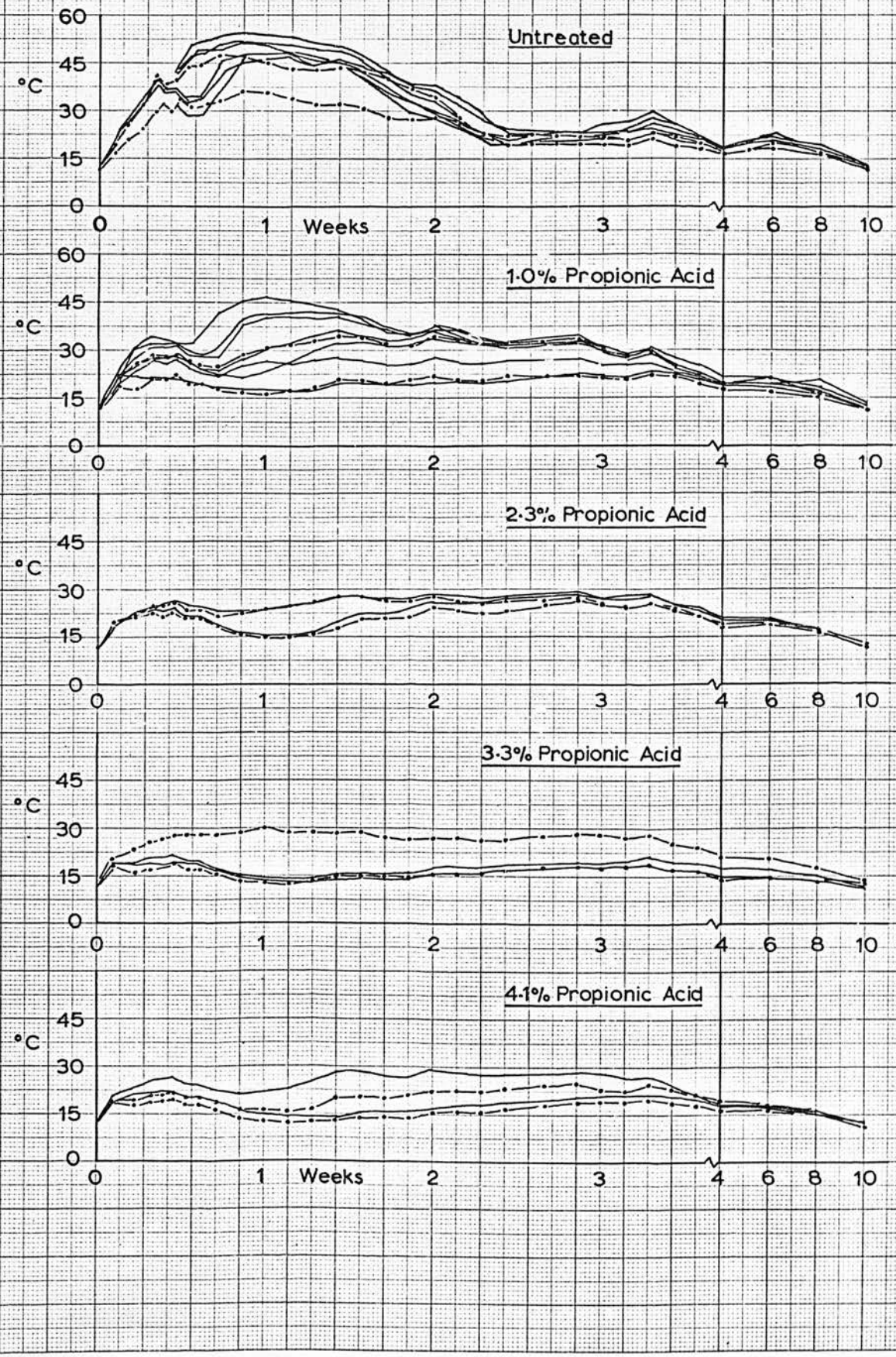


FIGURE 13 LOW MOISTURE HAY

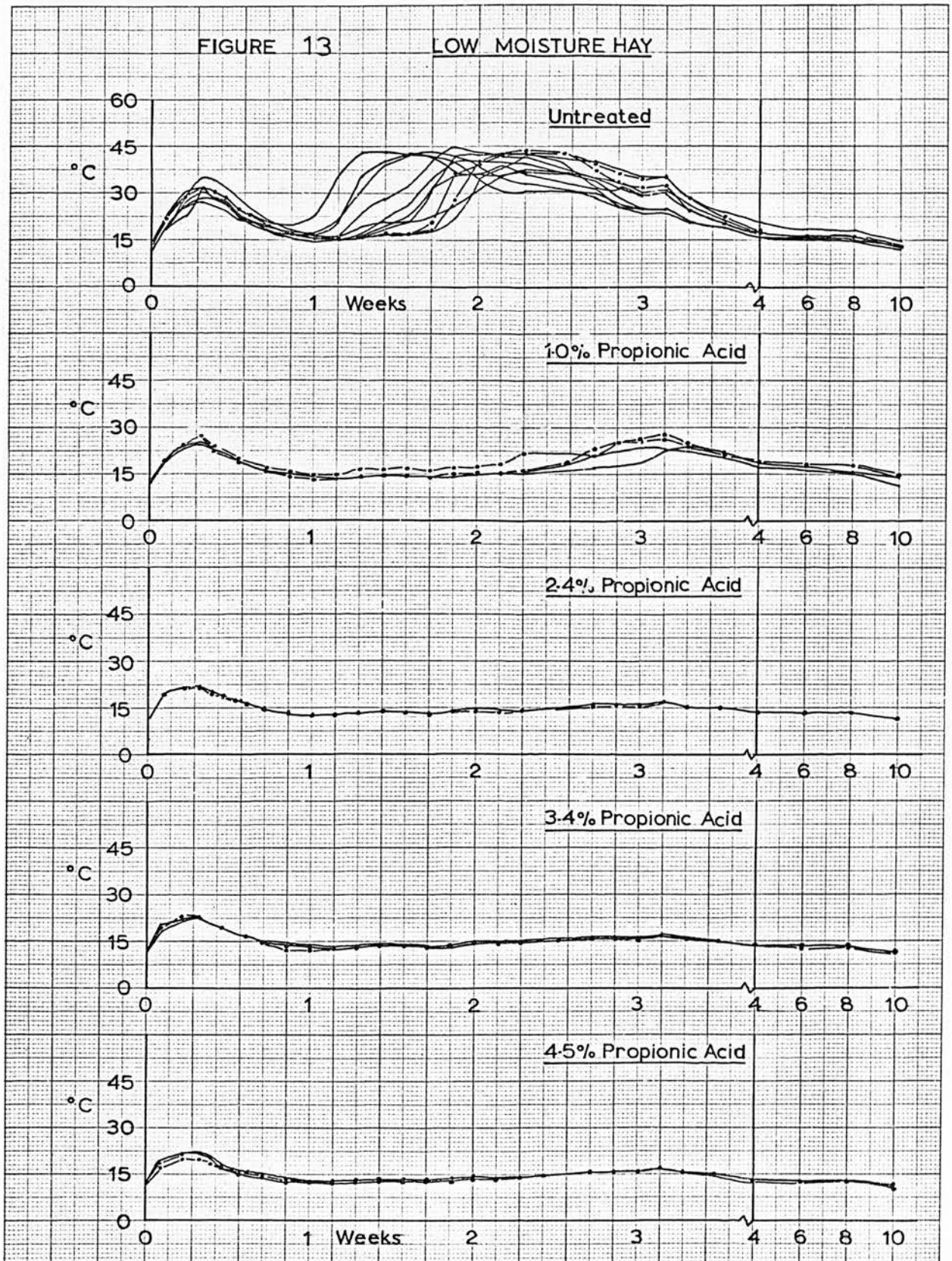
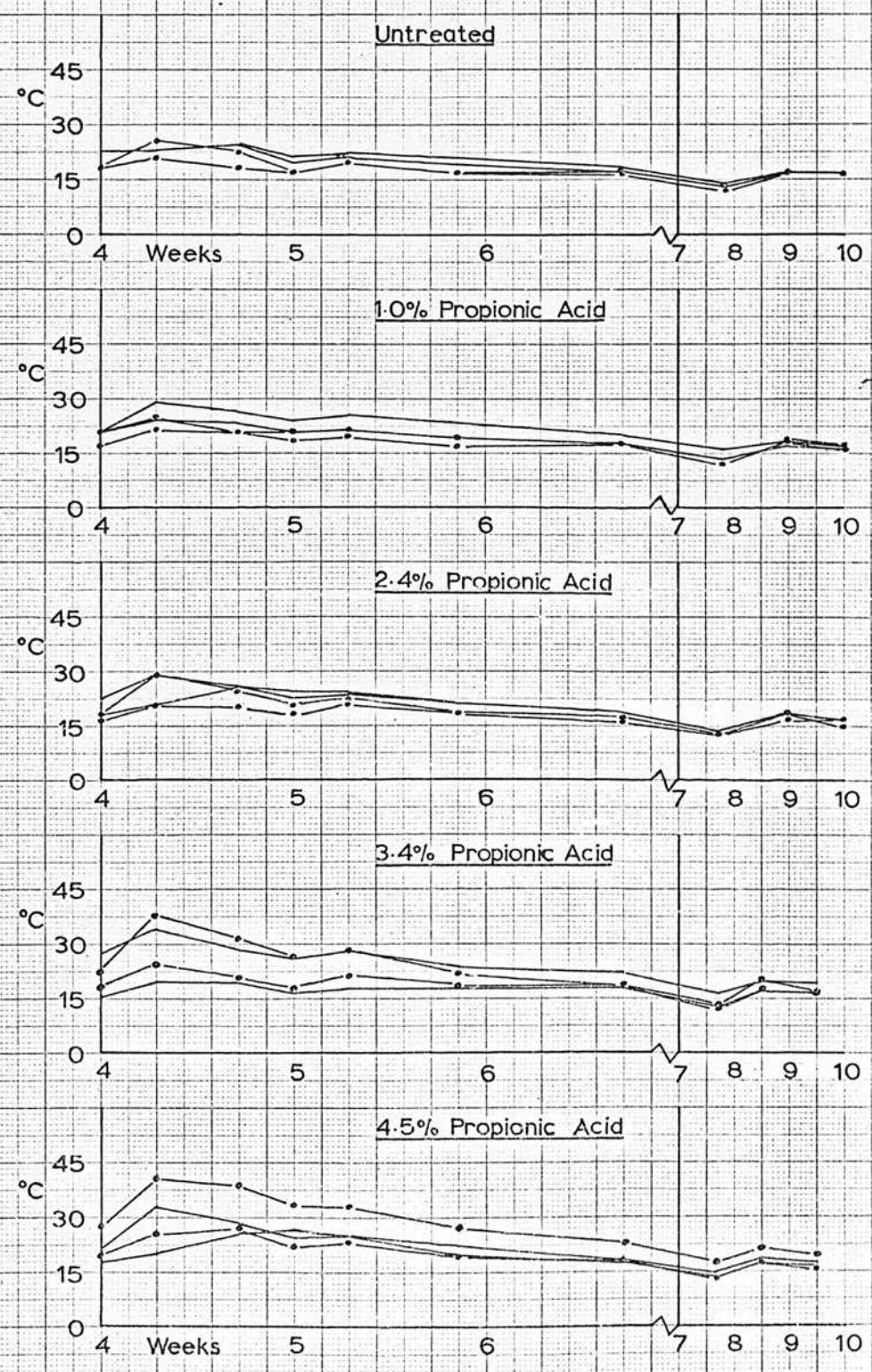


FIGURE 14 LOW MOISTURE HAY - FIELD CURED



not reach such high temperatures as the controls, but temperatures above the ambient were maintained for 3 to 4 weeks longer.

The temperature patterns followed by different bales within each acid treatment level showed considerable variation, this being most clearly seen in the high moisture hay treated with 2.1 per cent propionic acid. At day 5 the temperatures in different bales covered a range between 22° and 52°C. Fewer thermometers were used in batches at the other treatment levels, but a wide range of temperatures can still be observed.

In a few of the acid treated bales the temperature followed a pattern similar to that in the untreated bales in which an initial temperature peak was reached after 3 or 4 days followed by a drop of a few degrees and then a higher secondary peak, during the 5th or 6th days. Even at the lowest acid level, however, the maximum temperature was fully 20°C below that of the untreated bales. Those acid treated bales which did not show clearly defined initial and secondary peaks did still show a tendency to have more rapid heating taking place during days 1 and 2, and 5 and 6 than during days 3 and 4 (see 3.2 per cent propionic acid level).

While it is clear that all the acid batches had temperatures below those of the untreated batch, none of the treatments entirely eliminated heating. The 1.1, 2.1 and 3.2 per cent acid treatments progressively reduced the overall amount of heating, but the 3.5 per cent treated batches followed a temperature pattern almost identical to that in the 3.2 per cent batches.

b. Medium Moisture Hay (Figure 12)

Once again rapid heating took place in the untreated bales,

with an initial peak after 2 to 3 days. Subsequent heating took the temperatures to about 50°C, but as with the higher moisture batches, after 2 to 3 weeks the bales cooled down to the ambient temperature. Most of the bales followed a closely similar pattern.

In all the 1.0% acid treated bales an initial peak was evident at 2 to 3 days, although not as high as in the control bales. In some of these bales, however, no further heating took place, the bales following the ambient temperature, while in the others heating took place to varying extents and a maximum temperature of 47°C was recorded. In these bales temperatures above the ambient were maintained for 3 to 4 weeks.

The higher levels of acid treatment gave further control of heating and no temperatures above 30°C were recorded. At the 2.3% acid level slight heating up to 27°C took place during the 2nd week, after which temperatures declined to the ambient by the 8th week. At the two highest levels of acid one bale in each case showed heating to a maximum of about 30°C and maintained a temperature in this region for over 3 weeks. The other bales in these batches showed little or no heating with maximum temperatures in the region of 20°C.

c. Low Moisture Hay (Figure 13)

In the untreated hay the initial peak, reaching to over 30°C was followed by a delay of 4 to 10 days during which the temperatures declined almost to the ambient. A second period of heating then followed during which the temperatures rose to between 40° and 45°C. A gradual decline in temperature then took place until the ambient temperature was reached during the 4th week. As in the untreated

batches at the medium and high moisture levels, all the bales followed a similar temperature pattern.

During the first two days of storage all the acid treated bales heated to a temperature of about 22°C, and at the 2, 3 and 4 per cent acid levels no further heating took place, all the bales following identical temperature patterns very close to the ambient. At the 1 per cent acid level, however, slight heating took place during the latter part of the second week with temperatures of up to 28°C. It should be noted that this heating coincided with a slight rise in the ambient temperature and in the temperatures of the other batches.

d. Field Cured Bales (Figure 14)

The temperature of the field cured bales did not follow the same pattern in store as for the bales stored immediately after baling. In fact an entirely opposite trend seemed to take place in which the bales which had received the higher levels of acid had higher temperatures. The 1 per cent treated bales, for example, had a maximum temperature of 29°C while the 4 per cent treated bales reached 41°C. At the same time the corresponding batches which had been brought into storage immediately were at temperatures of 18°C and 14°C respectively. It should be remembered, however, that the temperatures of the bales were not monitored during the field curing period and heating in the untreated and possibly the treated batches could have followed its course before the bales were brought in at the end of four weeks in the field.

THE PATTERN OF CHEMICAL CHANGES

The results of the samplings carried out at 2 weeks and at

6 weeks together with the initial and final analyses help to build up a picture of the sequence of changes which took place in the bales. Figures 15, 16 and 17 show the changes in moisture content, water soluble carbohydrate (per cent in dry matter) and pH over the 12 to 14 week storage period.

a. Moisture Content (Figure 15A)

In the untreated hays loss of moisture did not take place at a constant rate throughout storage, but mainly during first two weeks, at the time when the highest temperatures occurred. By the end of storage the moisture content in all the untreated batches had fallen to between 15 and 20 per cent.

Increasing propionic acid levels progressively slowed down the rate of moisture loss, particularly during the first two weeks. This again may be correlated with the fact that the propionic acid treatments reduced the extent of heating. As already noted on page 57 the 2, 3 and 4 per cent acid treatments on the low moisture hay almost entirely eliminated the loss of moisture. At each of the acid levels the moisture contents of the high, medium and low moisture hays converged over the storage period until at the end of storage they were all nearly the same. The untreated hays, as already mentioned converged at about 17 per cent moisture content, the 1 per cent treated hays at about 20 per cent moisture, the 2 per cent hays at 25 per cent, the 3 per cent hays at 30 per cent and the 4 per cent hays at about 27 per cent moisture.

b. Water Soluble Carbohydrate (Figure 15B)

In the untreated hays the main loss of moisture content took place

during the first two weeks, with only slight further losses during the remaining 10 to 12 weeks. When the effects of acid treatment on the high moisture hay is considered it is seen that, at the 1 per cent acid level, the final water soluble carbohydrate content was much the same as for the untreated hay, but rate of loss was more gradual. The 2 and 3 per cent treatments reduced the rate of loss further so that the final water soluble carbohydrate content was, in these cases, higher. The 4% treatment (in fact 3.5% on the high moisture hay) follows a very similar pattern of water soluble carbohydrate changes to the 2 per cent acid treatment.

In the medium moisture hay during the 0 to 6 week period changes in the water soluble carbohydrate content followed a similar pattern to that in the high moisture hay, but at a slightly higher water soluble carbohydrate level. At the 1 per cent acid level the loss of water soluble carbohydrate continued very slowly during the second half of the storage period to finish at about 6 per cent in the dry matter. At the 2 and 3 per cent acid treatment levels, however, the final water soluble carbohydrate levels were fully 2 per cent higher at the end of storage than they were at the 6 week sampling.

In the low moisture hay the final water soluble carbohydrate content was not significantly different from the initial level at the 2, 3 and 4 per cent acid treatment levels. However Figure 15B shows that during the first 2 weeks the content dropped, but then during the next 4 weeks rose again after which it remained constant. At the 1 per cent acid level a similar but less marked trend was evident, although during the last 6 weeks the water soluble carbohydrate content dropped rather than remained constant.

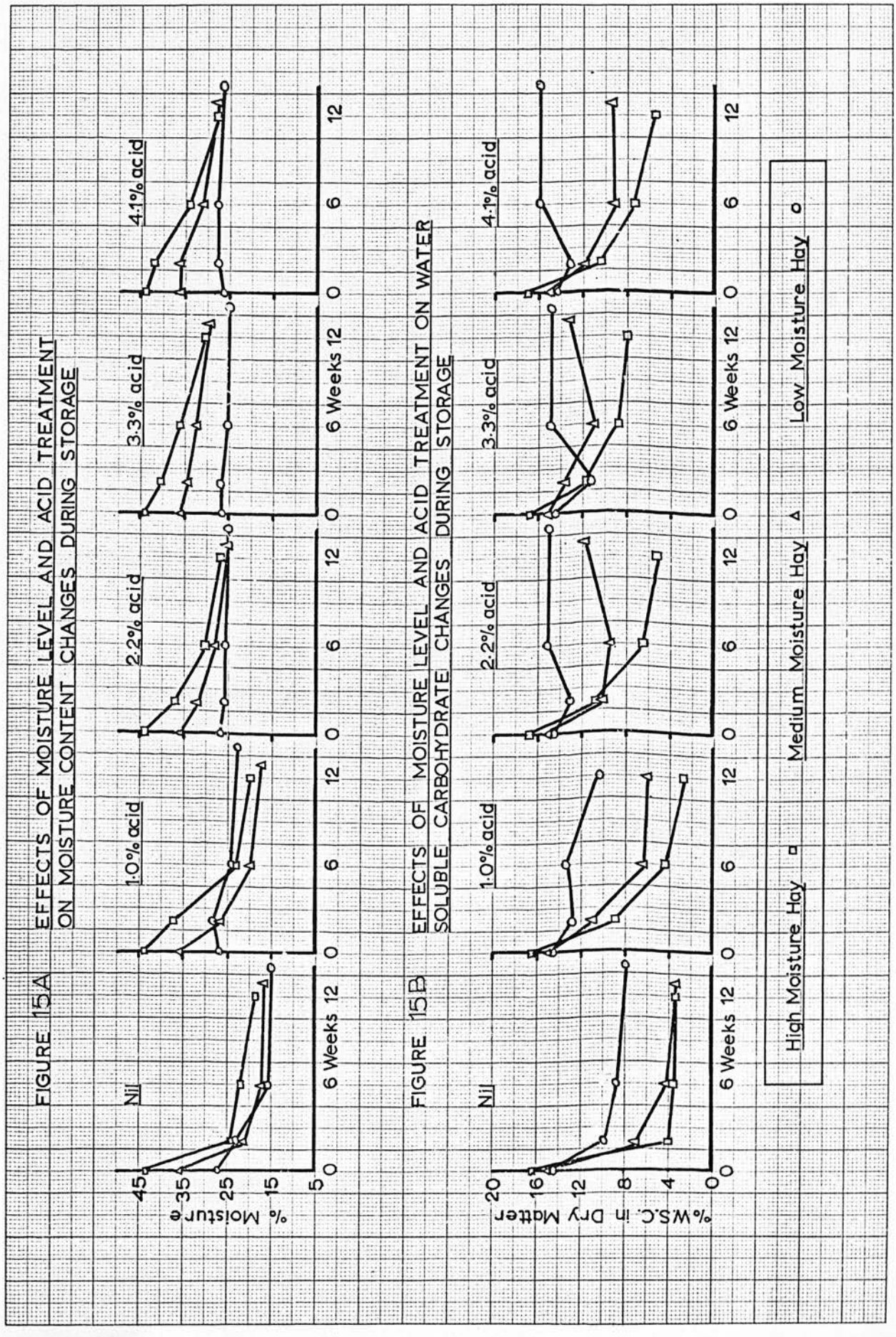
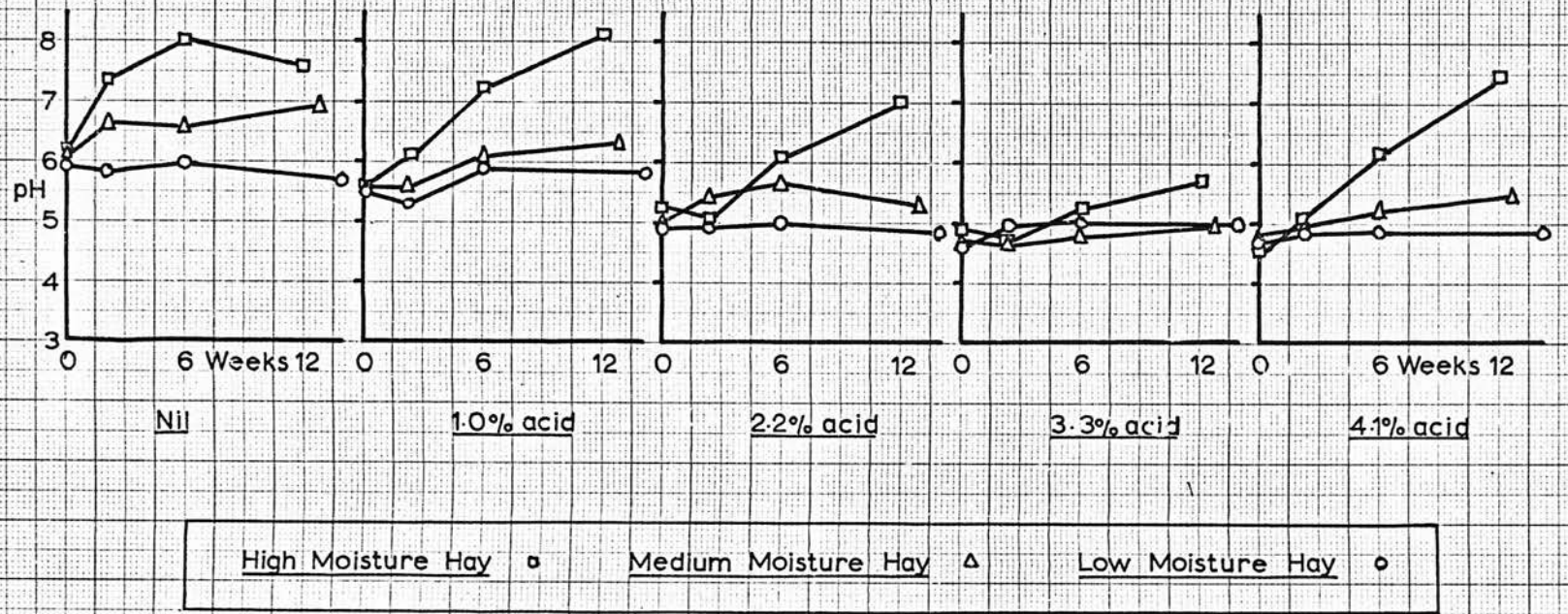


FIGURE 15A EFFECTS OF MOISTURE LEVEL AND ACID TREATMENT ON MOISTURE CONTENT CHANGES DURING STORAGE
Nil
1.0% acid
2.2% acid
3.3% acid
4.1% acid
% Moisture
45
35
25
15
5
0
6 Weeks 12
0
6
12
FIGURE 15B EFFECTS OF MOISTURE LEVEL AND ACID TREATMENT ON WATER SOLUBLE CARBOHYDRATE CHANGES DURING STORAGE
% W.S.C. in Dry Matter
20
16
12
8
4
0
High Moisture Hay □
Medium Moisture Hay △
Low Moisture Hay ○

FIGURE 16 EFFECTS OF ACID TREATMENT AND MOISTURE LEVEL ON pH CHANGES DURING STORAGE



c. pH Levels (Figure 16)

The series of graphs in Figure 16 show clearly the tendency for the pH of the high moisture hays to rise. In the untreated hay a pH of 8 was reached by the 6th week, declinining to 7.5 by the end of the storage period. The 1, 2, and 3 per cent acid treatments progressively reduced the rate at which the pH rose as well as lowering the initial pH of the hay. The high moisture hay which received 3.5 per cent acid (the '4' per cent level) had a pH rise as steep as that in the 1 per cent treated hay.

The rise in pH in the medium moisture hays was much less marked than in the high moisture hays, being less than 1 unit over the storage period in every case. In the low moisture hays pH changes were slight even in the untreated hay.

PATTERN OF MOULDING

a. Two week sampling (Table 9)

Extremely high counts of moulds and actinomycetes were found in the untreated high moisture hays. Mesophilic and thermophilic organisms formed a large proportion of those present (Malt salt agar, 37°C, Nutrient agar 55°C) reflecting the relatively high temperatures reached in these bales.

Much lower counts were recorded from the high moisture acid treated bales at the above incubation temperatures, but there were wide differences between the samples from the replicate batches. In the acid treated high moisture batches however, the counts of mesophilic moulds at 28°C (malt salt agar) were between 8 and 120 times higher than in the untreated hay. This trend was not repeated in the

TABLE 9 ctd.

Microbiological Counts — 2 wk sampling — FIRST Batch Experiment

HIGH MOISTURE

	MSA 28°	MSA 37°	NA 28°	NA 37°	NA 55°
0%	100,000	550,000,000	1,240,000,000	3,100,000,000	≥ 400,000,000,000
	-	-	-	-	-
1%	17,000,000	-	350,000,000	30,400,000	9,000,000
	830,000	860,000	1,180,000,000	1,220,000,000	54,000,000
2%	8,000,000	1,000,000	2,300,000	304,000	2,800,000
	11,200,000	3,400,000	4,700,000	3,900,000	9,800,000,000
3%	14,400,000	16,000,000	370,000,000	29,000,000	7,400,000,000
	2,600,000	8,000,000	10,400,000	1,700,000	108,000
4%	1,500,000	300	27,000,000	100,000	16,600,000
	10,400,000	6,000,000	27,000,000	11,000,000	2,800

TABLE 9 ctd.

Microbiological Counts — 2 wk sampling — FIRST Batch Experiment

MEDIUM MOISTURE

	MSA 28°	MSA 37°	NA 28°	NA 37°	NA 55°
0	40,000,000 5,200,000	20,000,000 4,400,000	11,000,000 56,000	4,600,000 43,000	6,400,000 37,000
1%	86,000,000 8,500,000	50,000,000 2,600,000	6,900,000 1,600,000	3,200,000 1,000,000	260 54,000
2%	5,900,000 9,900,000	5,300,000 5,000,000	95,000 9,600,000	51,000 170,000	101,000 1,230
3%	3,600,000 290	4,400,000 <10	24,000 143,000	21,000 10,000,000	14,000 2,010
4%	5,700,000 2,000	2,900,000 330	3,200,000 40,000	16,000 17,000	1,690 13,000

TABLE 9

Microbiological Counts — 2 wk sampling — FIRST Batch Experiment

LOW MOISTURE

	MSA 28°	MSA 37°	NA 28°	NA 37°	NA 55°
0	800,000	600,000	19,300,000	26,000	1,240
	1,800,000	1,600,000	100,000	25,000	540
1%	5,000	120	89,600,000	21,000	940
	400	120	26,000,000	67,000	780
2%	5,000	650	148,000	3,000	900
	150	<10	286,000	6,000	1,680
3%	200	<10	142,000	9,000	40
	40	<10	35,000	2,000	750
4%	100	<10	101,000	21,000	290
	100	<10	217,000	32,000	4,200

results for bacteria at this temperature (nutrient agar).

At the medium moisture level the 3 and 4 per cent acid treatments had counts of moulds and bacteria well below those of the untreated hays at all three incubation temperatures. Even in the untreated hays however the counts of these organisms were well below those in the high moisture hays. The 1 and 2 per cent acid treated medium moisture hays showed little or no tendency to have lower counts than the untreated hays at 28° and 37°C, although they did have fewer thermophilic organisms at 55°C.

The untreated hay at the low moisture level reached a maximum temperature of only 45°C and this is reflected in the low count for thermophilic actinomycetes on nutrient agar at 55°C. All the treated hays at this moisture content also had low counts at this temperature. The numbers of moulds at 37°C was very much reduced by the 1 and 2 per cent acid treatments and virtually eliminated by the 3 and 4 per cent treatments. At the lower temperature moulds were in slightly greater numbers but the same pattern emerged with only a few moulds being present at the higher acid levels. Bacteria did not seem to be inhibited to the same extent by the propionic acid and high counts were recorded in the 1 per cent acid treated hays. Counts at the 2, 3 and 4 per cent acid levels were not far below those of the untreated hay.

b. Assessment of Visible Moulding at the end of Storage

All the untreated bales at the high and medium moisture levels were completely mouldy, some, particularly at the high moisture level, showing severe browning. The acid treated bales at both of these

moisture levels were not uniformly mouldy, but had distinct and well defined patches of moulding. Table 10 shows the estimated proportion of moulding in the bales at each treatment level. This proportion decreased as the acid treatment level was increased at both moisture levels. At the medium moisture level 3 out of the 8 bales examined from the 3 and 4 per cent acid treatments did not show any visible moulding after thorough examination. On average, the 2 per cent acid treatment at the medium moisture level had the same extent of moulding as the high moisture hay treated with 4 per cent propionic acid.

Although not so clearly defined, moulding could be detected in the low moisture untreated and 1 per cent acid treated hays, but not in the 2, 3 and 4 per cent acid treated hays at this moisture content.

TABLE 10

Assessment of Visible Moulding

Acid Level	Bale Gradings	Total	Percentage Score
a) HIGH MOISTURE HAY			
0%	5 5 5 5	20	100
1%	3 4 4 2	13	65
2%	2 4 2 4	12	60
3%	1 5 1 3	10	50
4%	1 1 2 3	7	35
b) MEDIUM MOISTURE HAY			
0%	4 5 4 4	17	85
1%	4 3 3 2	12	60
2%	0 3 2 2	7	35
3%	1 0 0 3	4	20
4%	1 1 2 0	4	20

c) LOW MOISTURE HAY

Bales on which mould could be detected had a dusty appearance, but there were no clearly defined patches of mould growth as found on the bales at the higher moisture levels.

Untreated bales:- Dusty

1% acid level :- Dusty with growth of yellow mould

2%, 3% and 4% acid levels :- No visible moulding.

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c. Analysis of Selected Samples using the Mould Spore Dispenser

As in the two week sampling the sample from the high moisture untreated hay was particularly high in thermophilic actinomycetes, but counts of moulds from this sample were not as high as those from the mouldy parts of the acid treated bales, samples 7, 8 and 9. Sample 8, with moderate visible moulding had a very high count of thermophilic actinomycetes while the hay appearing to be the most severely moulded did in fact have very high counts of both moulds and bacteria at 28° and at 37°C (sample 9). The results are shown in Table 11.

Samples 2 and 4, which had been selected as having no visible moulding had the lowest counts of moulds, the differences between them and the mouldy samples 1, 3 and 5 being greater at the 28° than at the 37°C incubation temperature.

The mouldy samples from the medium and low moisture hays, except in the case of sample 3, had similar mould counts to those in the samples 7 and 8 with light and moderate moulding from the high moisture bales. The counts of bacteria and actinomycetes, however, were generally lower in the lower moisture hays.

The moulds most commonly isolated from the malt salt agar plates

TABLE 11

Counts of Microorganisms from Selected Hay Samples

(Using Air Displacement Sampler)

Sample Number	Moisture Level	Acid Treatments	Visible Moulding	Moulds		Bacteria & Actinos		
				Malt salt agar		Nutrient agar		
				27°C	36°C	27°C	36°C	55°C
1	Low	Nil	Dusty	28	7	51	93	16
2	Low	3%	None	5	5	20	6	5
3	Medium	Nil	Severe	6	9	164	47	14
4	Medium	2%	None	4	3	16	13	3
5	Medium	2%	Moderate	57	2	42	13	1
6	High	Nil	Severe	4	13	236	127	>2000
7	High	3%	Light	28	13	139	196	26
8	High	3%	Moderate	32	21	405	157	>2700
9	High	3%	Severe	282	392	>3000	>3000	414

were of the Aspergillus glacus group although Penicillium species were also prevalent. The high counts on the nutrient agar plates were predominantly of white actinomycete species although many other types of colonies were observed including those of spore forming bacteria and yeasts. In addition, the fungi Mucor and Rhizopus were frequently found spreading across the nutrient agar plates.

PROPIONIC ACID DISTRIBUTION WITHIN THE BALES

The following results were obtained from the analysis of 12 cored samples taken from a bale at the low moisture level which had received the 2 per cent acid treatment. As shown in Table 1, the bales at this acid level were calculated to have received, in fact, approximately 2.4 per cent acid. The samples were taken from sections B and D (Figure 17), which it was hoped would be free from end effects, and parts 2, 3, 6, 7, 10 and 11 were taken in each case. The analyses were carried out by BP Chemicals (International) Ltd. using the Steam Distillation technique. The samples were taken from the bale after the 3 month storage period. No visible moulding was present on the bale.

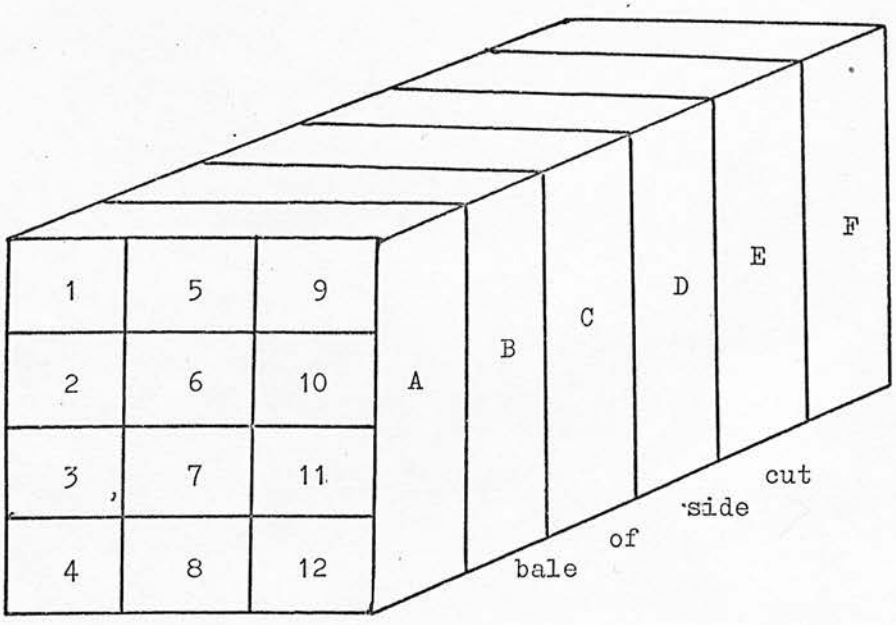
The results show that the distribution of the acid within the bale was not uniform and all the parts of section D received a higher level of acid than the corresponding parts of section B. Some variation also took place in the distribution of acid within each section of the bale.

The overall mean acid level was very near to the application rate of 2.4 per cent which was calculated shortly after baling.

FIGURE 17

FIRST BATCH EXPERIMENT

Location of Samples Analysed for Propionic acid



Although only the result from a single bale, it might be taken to indicate that, when little or no moulding takes place, the propionic acid in the bale is not lost during a prolonged storage period.

TABLE 12

Propionic Acid Distribution

Sample	Per cent Propionic Acid		
	Section B	Section D	Mean
2	2.26	3.90	3.01
3	2.14	3.73	
6	1.81	2.34	1.95
7	1.19	2.46	
10	1.95	2.32	2.12
11	2.08	2.14	
Mean	1.91	2.82	2.36

FIRST BATCH EXPERIMENT

DISCUSSION

The biological changes taking place in hay after baling are complex and in this experiment no one of the parameters measured during the course of the experiment could, on its own, give a comprehensive picture of the changes.

Direct counts of the microorganism population, for example, should give the most direct assessment of biological activity, but in practice the errors involved during the sampling of the hay and the quantifying of the population make the results of limited value. There is also the fact that the population of the various organisms taken at the end of a storage period will not yield much information on the changing balance of these organisms during earlier stages.

Perhaps the most easily measured barometer of biological activity is the temperature pattern. Even this, however, gives a biased picture for the temperature rise is not just a reflection of the biological heat output, but is also influenced by the specific heat capacity of the material and the rate of heat loss. In the bales the former will change as the moisture content of the hay fluctuates, while the latter will depend upon the surface area of the bale and on the ambient temperature. Thus, for bales of the same dimensions, a bale with a high moisture content will require a greater heat output to raise it through the same temperature rise than a bale of lower moisture content.

In this experiment therefore, a bale from the medium moisture hay which reached the same temperature as a bale from the high

moisture hay will in fact have had a slower rate of heat output than the high moisture bale. The results show, however, that the moisture content itself is not static but falls due to the loss of water by evaporation. Eventually, in untreated hay, it is the moisture content which limits further biological activity when it falls below the level at which the microorganisms can live. The temperature patterns must therefore be viewed in the light of the differing initial moisture contents, and therefore specific heat capacities, of the hays and the changes in moisture contents during storage.

Nevertheless the temperature patterns do give a very useful means of comparing the biological activity which took place in the bales of the different treatments. The temperature readings were of particular value because they gave the first indication of any biological activity, and when taken frequently, gave the best indication of the fluctuations in the biological activity.

The periodic samplings for chemical analyses, on the other hand, gave a picture of the cumulative effect of the biological activity and enabled estimates to be made of the groups of organisms giving rise to the biological activity.

The results of the experiment show clearly that the application of propionic acid significantly reduced the level of biological activity in hay over a range of moisture contents, leading to a reduction in dry matter and water soluble carbohydrate losses and a decrease in the extent of moulding.

In the low moisture hays, with an initial moisture content of about 27 per cent, the 2, 3 and 4 per cent treatment levels prevented virtually all moulding as indicated by the very low mould counts on the samples taken at 2 weeks, and in the corresponding sample examined

at the end of storage and by the mould free appearance of the bales. Heating was almost entirely eliminated from these bales except during the first few days, when respiration of plant cells was likely to have been the source. It might have been expected that this too would have been eliminated by the acid, as contact with the acid will cause rapid plasmolysis and death of cells. It is possible that isolated pockets of untreated hay could have given rise to this heating.

This initial heating was, however, reduced by the acid treatments when comparison is made with the untreated hay, and was not sufficient to bring about any significant loss of moisture or water soluble carbohydrate.

The untreated low moisture bales became dry and dusty by the end of the storage period due to the growth of moulds, and quite extensive heating took place. The moulds present on the hay were of the Aspergillus glaucus group and had a yellow appearance due to the massive number of yellow perithecium which were formed and which could be shaken from the hay to form a deposit. In the hay which received the 1 per cent acid treatment the extent of heating and moulding was reduced. Some moulding was present, however, and the losses of water soluble carbohydrate and dry matter were much closer to those of the untreated hay than to the hay which received the 2 per cent treatment. The onset of the moulding nonetheless was delayed by the 1 per cent propionic acid treatment. A similar delaying effect has been noted by other workers (Lewis, 1951; NAAS, 1969).

The fact that the moulding was coincident with a rise in the ambient temperature would suggest that the propionic acid had

inhibited, but not entirely prevented mould growth and that it needed the warmer ambient temperature to increase the growth rate to the acceleration stage at which heating would take place. The picture is further complicated by the strong possibility that the acid distribution was not even throughout the bales. Although dry matter and water soluble carbohydrate losses in the 1 per cent treated hay were nearer to those of the untreated hay than of the 2 per cent hay, the temperature was nearer the pattern of the 2 per cent than the untreated. This would confirm the previous suggestion that slow moulding was taking place in the 1 per cent hay but not at a rate fast enough to produce heating until encouraged by the warmer ambient temperature.

In a number of cases, particularly at the three highest acid levels, increases in the water soluble carbohydrate content of the treated hay were recorded. Slight increases have been found by other workers with dried samples of hay*, but it is difficult to see how rises of the magnitude of those of the present experiment could have occurred, particularly as they only occurred during the 2 to 4 week period, and did not continue during the rest of the storage. Fructose standards were used throughout in the analysis procedure and so it was unlikely to have been a fault in the method which gave this increase. It is possible that sugar increases could arise from the breakdown of hemicelluloses and if this took place the presence of the breakdown products xylose, galactose and arabinose would be expected. However, when the sugars from the hay samples which had increased in water soluble carbohydrate were separated by paper chromatography, no trace of these breakdown products was found. Similar increases in water soluble carbohydrate have been recorded in silage experiments where an additive such as a formalin/formic acid mixture has prevented

* Greenhill *et al*, 1961.

biological activity in the silage, (Mc Donald* 1974), and it has been thought that cellulases might have been active. In the medium moisture hay slight increases in water soluble carbohydrate were also recorded. These took place at the 2 per cent and 3 per cent acid levels between the 6 and 8 week samplings.

The position in the medium and high moisture hays was not so clear cut as in the low moisture hay and considerable moulding was found in bales at all the acid levels. However, it was difficult to provide a really reliable measure of the level of mould contamination. Where bales are made up of both mouldy and mould free parts, core samples will contain a mixture from these parts. For chemical analysis this will give a mean result which is a true mean, but for mould counts this is not the case. The parts of the sample which are mouldy contain such a large number of mould spores, usually measured on a log scale (10^6, 10^7 etc) that to take a mean between this and a part with, say, 100 or 200 spores per gram gives a result which is heavily weighted towards the count of the mouldy part. Thus although the results in Table 9 do follow a general trend of decreasing counts with higher acid levels, the counts do not necessarily give a true picture of the relative mould contamination. The method of sampling could also partly account for the considerable variation found between counts of different replicates of the same treatment. It would seem likely, for example, that the low mould counts in the 3 per cent medium moisture hay, replicate 2, would have occurred because the corer did not pass through a mouldy patch. The conclusion could not be drawn that the bale did not contain any mouldy parts.

As the main reason for the use of propionic acid is to eliminate the presence of mould, and mouldy patches, in the bales, the use of

* and Henderson

core samples was therefore considered to be of limited value, and it was thought that a visual assessment of the extent of moulding at the end of storage, along with a more detailed examination of parts of the bales, would yield further useful information.

The serial dilution method used for determining mould counts on the 2 week samples was found to be more time consuming than could be justified by the results obtained and was replaced by the method in which mould spores and bacteria were displaced from a glass jar containing the sample of hay.

The relatively high level of microbiological contamination found on the high moisture hay samples at 2 weeks was confirmed in the samples at the end of storage, where even the sample which was considered by visual examination to be least contaminated had a reasonably high level of moulds when incubated at 27°C.

It is also of interest to note the effect of acid treatments on the balance of microorganism counts at the different incubation temperatures. At 2 weeks the high moisture untreated hay had very high counts of thermophilic actinomycetes, but low counts of moulds at 27°C which would have been discouraged by the high temperatures. A large number of bacteria, mainly actinomycetes, however, were isolated at 27°C. This position was also found in the high moisture, untreated sample at the end of storage. In the acid treated high moisture hays the same high temperatures were not reached and so the low temperature moulds were not inhibited. Thus higher counts for moulds isolated at 27°C were found in the treated than in the untreated high moisture hay at both 2 weeks and at the end of storage. The counts of actinomycetes at 27°C at 2 weeks were slightly lower in the

acid treated hays, but at the end of storage were very high in the acid treated hay which was classified as being severely mouldy. The high moisture treated hays with light and moderate moulding had counts similar to those in the untreated hay. The hay sample classified visually as 'medium mouldy' in fact contained the highest level of thermophilic actinomycetes (55°C) and must have reached higher temperatures than the 'very mouldy' sample. The 'medium mouldy' sample most nearly resembled the untreated hay in the microorganism counts.

Similarly, in the medium moisture hay at the end of storage the 'mouldy' sample of treated hay had a higher count of moulds at 27°C than the corresponding untreated sample, which, on the other hand, had more moulds at 36°C. This trend may also be seen to some extent in the 2 week sampling between the 1 per cent treated and the untreated hays. As would be expected it was the low moisture acid treated hay which had the lowest overall counts of microorganisms at 2 weeks and at the final sampling.

The visual assessments clearly demonstrated the nature of the problem when it comes to complete preservation of the hay. In the high moisture hay none of the bales were completely mould free while in the medium moisture hay only a few bales were free of mould. The considerable variation in the amount of mould between bales at any treatment level corresponds with the temperature results in which widely differing temperatures were recorded for different bales of the same treatment (Figures 11 and 12). The variation in acid distribution is likely to have been partly responsible for this, but variations in moisture content within and between bales may have been present and contributed towards the presence of mouldy patches.

The question of the real extent of moisture and acid variations within the bales is a vital one, which gave rise to the distribution experiments carried out in the two years following this first experiment and described in section 3.

It remains to give some comment on the seemingly anomalous results in which the 4 per cent treatment in the high and medium moisture hays were not better preserved than the 3 per cent hays, and, in some cases were less well preserved. This tendency may be observed in Tables 2 to 8. In the high moisture hay, while the moisture level of the 4 per cent treated hay was little different from that of the 3 per cent hay, the estimated level of acid treatment was only 3.5 per cent for the '4 per cent' hay while it was 3.3 per cent for the '3 per cent' hay. It would have been surprising, therefore, if there had been a significant improvement with the higher acid level.

In the medium moisture hay it will be seen from table 1 that the initial moisture content of the 4 per cent treated hay was over 6 per cent higher than that of the hay which received the 3 per cent treatment. This high moisture content may have been sufficient to outweigh the 0.8 per cent difference in the actual acid levels applied and lead to the greater levels of deterioration reflected in the chemical and microbiological analyses. Although the visual assessment of moulding at the two acid levels was the same on average (Table 10), the 3 per cent treated hay had two bales free of mould whereas the 4 per cent level gave only one bale free from mould thus adding support to the results of the other analyses. Only the dry matter losses show an opposite trend, a result which is difficult to explain unless the sampling or analysis was inaccurate.

It is of interest to note that when acid was applied to the hay passing through the baler, the bales became less dense. This was presumably due to the 'lubricating' effect while the surface of the hay was still wet with the acid. Various experimentors have demonstrated that density can effect the deterioration of hay* and it is possible that the reduced density would aid the acid in reducing the temperatures. At the lower density air can move more freely in the bale which also has a greater surface to weight of hay ratio. Thus the heat produced by a smaller weight of hay is lost more rapidly from the relatively greater surface area and the rise in temperature in the bale is reduced.

While it is not possible to tell the effect of the density variation on this experiment, the variation of density found in this experiment would only be expected to reduce the temperature by a few degrees, this affecting only the difference between the untreated and the treated batches as a whole, and not the difference between the various acid treatments.

The field curing period of 4 weeks generally had a deleterious effect on the quality of the hay which was most marked in the case of the acid treated hay. The field curing did not seem to help the drying out even in the untreated hay, while the parts of the bales which became damp with rainwater running off the plastic cover became mouldy even in the bales with the highest acid treatment. Considerable heating took place in these acid treated bales after being brought into store and it would seem from the graph in Figure 14 that increasing acid levels gave rise to higher temperatures in the hay. However, it should be remembered that the effect of propionic acid can be to delay the onset of moulding and heating. It is likely, therefore, that at the lower acid levels heating took place while the hay was

* Nelson, 1966, 1968 & 1972; Hopkins et al, 1954.

still in the field, but at the higher acid levels, of 3 and 4 per cent, heating was delayed until the 4 to 6 week period giving rise to the relatively high temperatures shown in the graphs.

It was hoped that out of this experiment it might have been possible to suggest levels of propionic acid which a farmer might apply to his hay over a range of moisture contents to preserve it effectively. Certainly in the low moisture hay the 2 per cent and higher levels were clearly adequate to preserve this hay of about 27 per cent moisture content, while the 1 per cent level was inadequate. In the medium and high moisture hay, however, no clear dividing line between adequate and inadequate levels could be drawn. While in the medium moisture hay some of the bales in the 3 per cent and 4 per cent treatments were free from patches of mould, other bales in the same batches did contain mould. It is possible that with improved distribution of the acid complete preservation might have been obtained in this medium moisture hay.

In the high moisture hay results were less hopeful and it would seem that moisture contents of over 40 per cent complete preservation could not be obtained without very much better distribution and possibly a more even moisture content. Better distribution may not only be a question of applicator design, but also of the profile of the swath as it enters the baler. From plate 1 it can be seen how variable the swath is at the time of baling and there is little doubt that a more even swath would make acid application less of a problem.

Although providing a valuable comparison of the treatments in this experiment, the small batches containing 9 bales may not give results representative of those that might be expected from bigger

bulks of hay such as might be used commercially. Thus the need was felt to extend the experimental work to bigger batches of, say, 50 bales. At the same time this kind of experiment could not furnish information on the levels of acid needed to preserve hay when the acid is applied evenly to hay of an even moisture content. The first issue is dealt with in the second batch experiment, while the second is taken up in the mini-bale experiments described in Section 2.

SECOND BATCH EXPERIMENT

AIMS

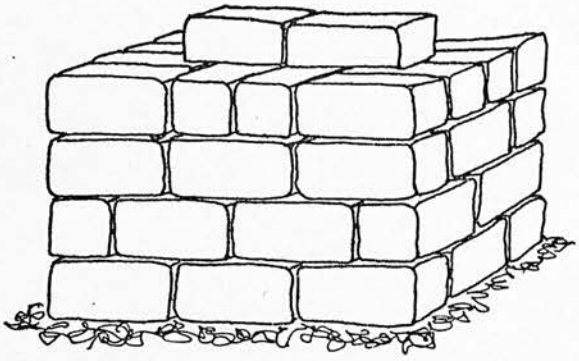
This experiment was carried out in order to establish whether the conclusions from the first batch experiment, in which 9-bale batches were used, held good for much larger batches of hay of the kind that a farmer might build in a hay shed. In the first experiment none of the acid treatments applied to the high moisture hay provided effective control of the moulding and so in this experiment the batches were only made in the medium moisture (30 to 40 per cent moisture) and low moisture (20 to 30 per cent moisture) ranges. More emphasis was placed on the low moisture range as this appeared from the first experiment to be the level at which the propionic acid treatment could be most successful using the present techniques.

In this low moisture range hay at between 25 and 30 per cent moisture was aimed at with treatments of 2, 3 and 5 per cent propionic acid to be applied, while at the higher moisture range of 35 to 40 per cent the 2 and 5 per cent acid levels were to be applied. Untreated control batches were to be made only at the low moisture level. In view of the greater variation expected within the larger batches three replicates of each treatment were planned to be arranged during storage in three randomised blocks as shown in Figure 18C

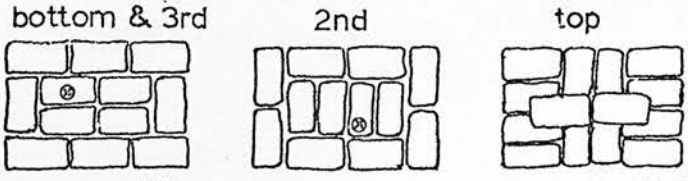
A batch size of 50 bales was chosen as being a suitable compromise between the essential requirements of having large batches with replication, and on the other hand a limited quantity of hay, and storage space available, with a limited supply of labour to handle, weigh and sample the bales. The 50 bale batches could conveniently be fitted

Figure 18

a) Design of the 50 Bale Batches

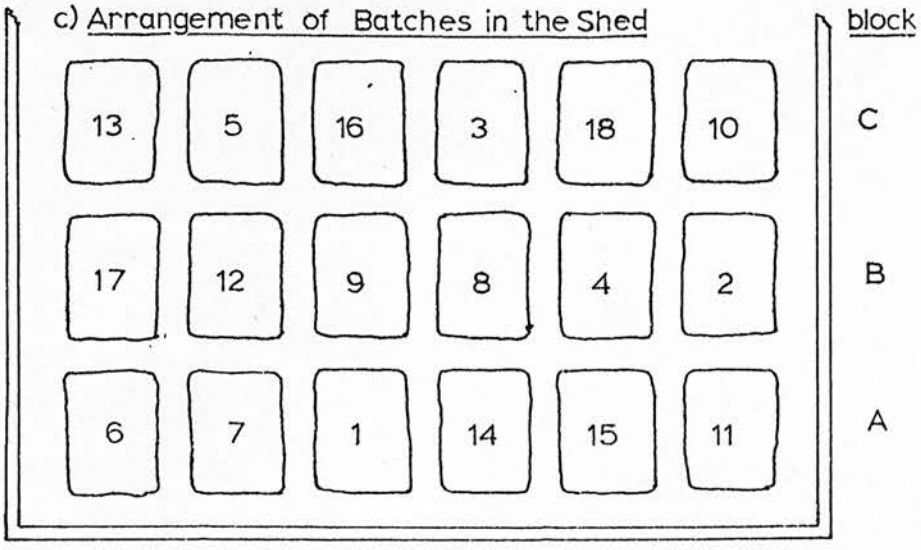


b) Plans of the Separate Layers



⊗ positions of thermometers in the 2nd & 3rd layers

c) Arrangement of Batches in the Shed



into one trailer load for weighing, and sampled efficiently using the extendable corer designed by Alexander et al (1969).

The use of such large batches, however, imposed other limitations on the design, particularly with regard to the controls. To allow 150 bales at each moisture level to go completely mouldy would have been not only expensive and a waste, but would have posed considerable problems in disposal. At the higher moisture level untreated hay would become both unpalatable and possibly toxic as animal feed and dangerous to anyone handling the hay due to the likelihood of thermophilic actinomycetes being present, the causal organisms of farmer's lung disease. In view of these considerations it was decided to make untreated control batches at the low moisture level only, and to limit the control to two batches only, with a third batch being barn dried for comparative purposes. Although it was realised that this would limit the scope of the statistical analysis of the results it was expected that reliable conclusions would still be able to be drawn.

METHODS

Field and Storage Work

During early July 1971 hay was made from 8 acres of a 3rd year ley largely comprising ryegrass and red clover which was cut using a rotary mower. Tedding and turning were commenced on the day of cutting and carried out in accordance with the established techniques of fast haymaking. Immediately prior to baling, the hay was windrowed by running two swaths into one, which was then of a suitable size for baling.

As in the first batch experiment frequent moisture determinations

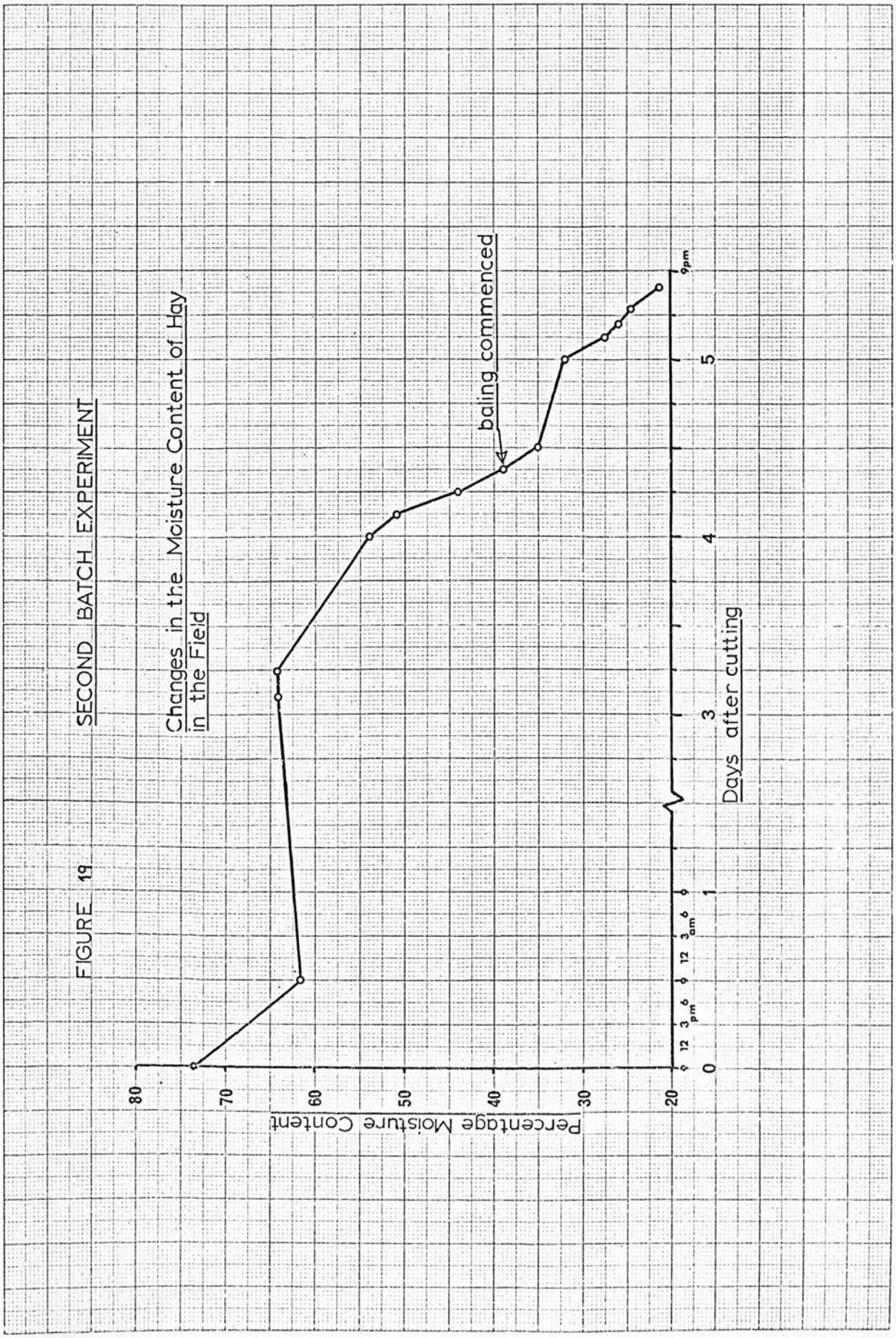
of the hay as it dried in the field enabled preparations to be made for baling as the desired moisture contents were approached. Figure 19 shows that during the first three days after cutting very little drying took place, the weather being dull and wet, but during the latter part of the 4th day, and during the 5th and 6th days ideal haymaking conditions occurred and very rapid, in fact from the point of view of the experiment too rapid, drying took place. The high moisture batches were baled during the 5th day after cutting, and the low moisture batches on the 6th day.

For each batch of 50 bales the baling of 2 or three swaths the length of the field was required. Adjacent swaths however varied in the quantity of hay in them, which could therefore influence the propionic acid treatment level being received by the hay. Rather than try to select similar swaths to be used to make each individual batch, which would have been difficult in practice, the swaths for each batch were chosen randomly from across the field so that the variation was incorporated into the batches randomly, rather than eliminated.

Immediately after baling, each batch was loaded onto a trailer and weighed at a weighbridge together with the tractor and the trailer. After the bales had been unloaded in the shed the tractor and trailer were reweighed so that the weight of the batch could be obtained. The weighbridge was accurate to the nearest 7 lbs. The batches were all built according to the design shown in Figure 18a and arranged in the shed as shown in the same figure. The same shed as in the first batch experiment was used.

The batch to be barn dried was weighed as with the other batches, labelled and then built into a stack along with other bales which were

FIGURE 19 SECOND BATCH EXPERIMENT



not part of the experimental work, but which were from the same field and were baled at the same time. The bales were dried by means of cold air ventilation powered by a diesel engined Lister fan. When drying was complete and the stack dismantled the labelled bales were recovered and taken to the shed to be built into their 50 bale stack along with the other batches.

At the end of the three month storage period and before the final sampling, the batches were, in turn, loaded onto a trailer and weighed as at the start of the experiment.

Experimental Methods

Sampling

Sampling of the hay in the field prior to and during baling was carried out in the same way as in the first batch experiment and described on page 40.

The batches of hay were sampled 2 weeks and 6 weeks after baling using an extendable corer capable of penetrating right through the stack (Alexander et al, 1969). This design of corer was found to be more efficient than the design used in the first batch experiment because it could be kept sharp much more easily. Supplied with the corer was a special tool which trimmed the cutting edge and kept it sharp but slightly serrated. This type of cutting edge cut very easily through the hay so that a hand powered woodworker's brace could be used to turn it rather than an electric drill.

A single core passing diagonally from top to bottom of the stack through at least 5 bales, was taken at each sampling. These samples contained about 1 kg of hay. At the end of the 3 month storage period

cores were taken individually from each bale in the same way as in the first batch experiment (Figure 5) and bulked to form a more representative sample of the whole batch. This sample was subdivided for chemical analysis and moisture determination.

Propionic acid Application

The propionic acid applicator used in this experiment was very much simpler than the one used in the first batch experiment, but was fitted to the baler in a similar way. A full description of this 'low pressure' applicator and of the other applicators used in the course of the experimental work are described in Appendix 7.

The acid was pumped by means of an electrically driven centrifugal pump from a 5 gallon plastic container on top of the baler to a spray boom positioned horizontally above and between the pick-up reel and cross auger of the baler (Figure 20). Pointing downwards from the ends of the spray boom were two spray nozzles which delivered the acid in two slightly overlapping fans across the width of the baler aperture. A number of different sizes of nozzles could be fitted to the spray boom thus providing a range of rates at which the acid could be applied. The exact flow rate through each size of nozzle had been established by BP Chemicals (International) Ltd., who also supplied the applicator and gave technical assistance during its operation.

As in the first batch experiment the applicator could only be set once the rate of baling of the hay was known. The following procedure was therefore used:

1. The rate of baling was estimated by timing the production of 5 or 6 bales, weighing these bales, and calculating the rate

Figure 20

LOW PRESSURE APPLICATOR MARK I

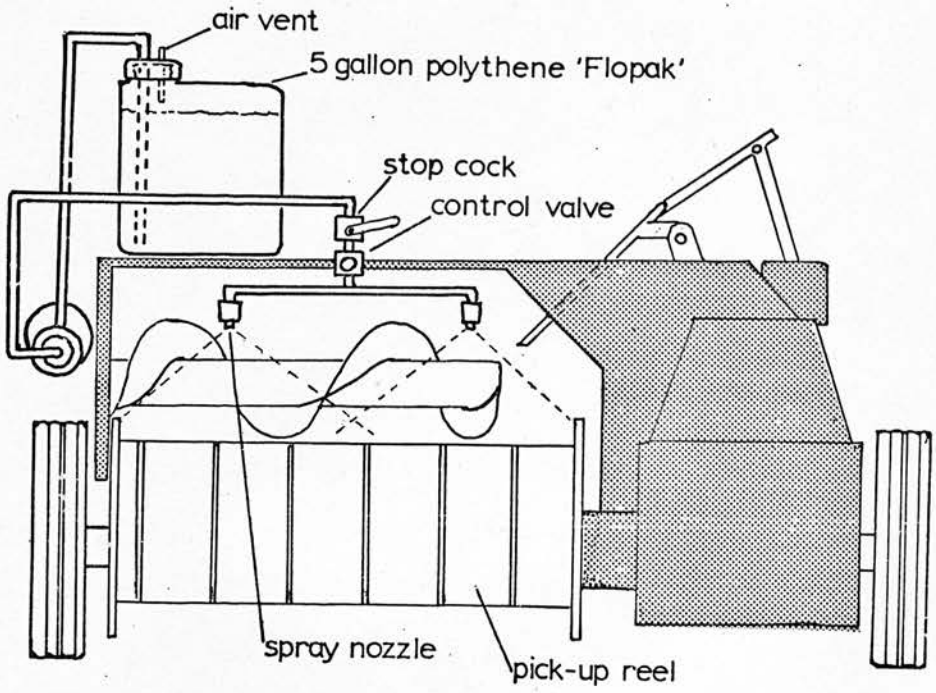


Diagram 1 Front view

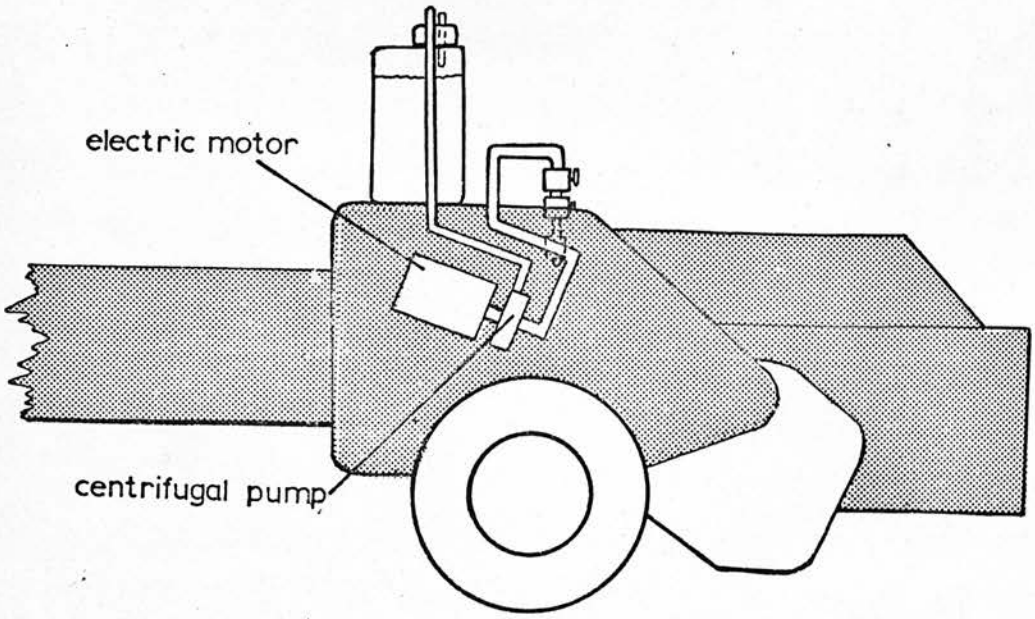


Diagram 2 Side view

in kilograms of hay per minute.

2. From the rate of baling the rate of flow of acid for the appropriate acid level was calculated.

3. The size of nozzle providing most nearly the correct acid flow rate was selected and the nozzles fitted to the spray bar.

By using different tractor speeds together with the range of flow rates provided by the different nozzles all the desired levels of acid treatment could be applied.

A note was kept of the flow rate being used for each batch and the rate of baling was checked by timing the production of, and then weighing, a number of bales. From this information the actual level of propionic acid being received by the hay could be calculated.

Temperature Readings

Temperature readings were taken at two locations in each stack. Mercury thermometers in tubes as shown in Figure 6 were inserted from the top into a bale of the third layer and from the side into a bale of the second layer at the positions shown in Figure 18b. Readings were taken daily during the first few weeks of the experiment and then less frequently as the temperatures neared the ambient.

Chemical analyses and Moisture Determinations

Moisture determinations were carried out using the oven drying and quick drying techniques described in the first batch experiment on page 47. Water soluble carbohydrate and pH determinations were carried out on the 2 week, 6 week and final samples as well as the samples taken at the time

of baling in the same way as in the first batch experiment.

Microbiological Analyses

The air displacement sampler described in the first batch experiment on page 50 was used to compare the levels of mould and other microbiological contamination on the core samples taken at 2 and 6 weeks after baling. Two separate subsamples of each batch sample were examined and plates of malt salt agar and nutrient agar exposed and incubated at 27°C and 36°C for both subsamples. The malt salt agar is selective for the growth of moulds and the nutrient agar is suitable primarily for the growth of bacteria and actinomycetes. Nutrient agar plates were also exposed and incubated at 55°C in order to disclose the presence of any thermophilic actinomycetes.

The final samples taken at the end of the storage period were not examined in this way, but a visual assessment of moulding was carried out on bales from one replicate batch at each treatment level. Four bales from each of these batches were randomly selected. Each bale was split into five sections of roughly equal size and an assessment made of the extent of moulding visible on the four exposed cross-sections of the bale. A grid divided into 16 sections was placed over each cross-section in order to aid the quantitative assessment of the moulding and the drawing of any mould patches which were present (Figure 21).

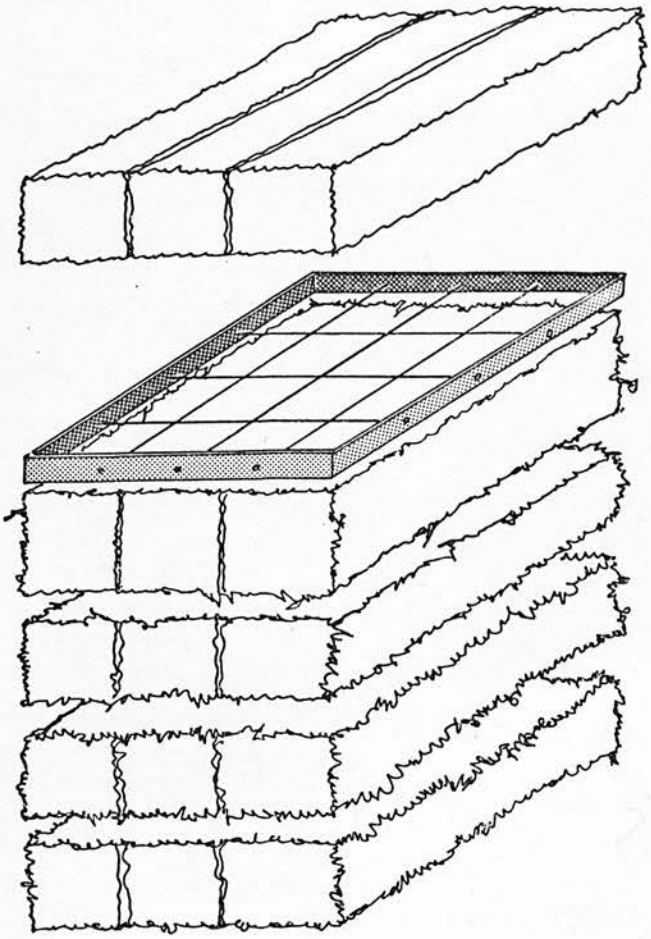


FIGURE 21

USING A GRID TO ASSES VISIBLE MOULDING
IN SECOND BATCH EXPERIMENT

RESULTS

The first batches to be baled in both the high moisture and low moisture ranges, batches 1 and 7 respectively, had initial moisture contents within the intended ranges. The rapid drying of the hay in the swaths due to the warm dry conditions, however, resulted on both days in a tendency for subsequent batches to have lower and lower moisture contents, as will be seen from Table 14. Replicate batches at each treatment level therefore tended to differ quite widely in their initial moisture content. The moisture contents of the low moisture batches were lower than intended except in the control batches which were baled first. The batch which was to be barn dried had almost fallen to the safe moisture level of 20 per cent before baling took place.

The acid treatments also turned out to be quite widely different on replicate batches at the same intended level, particularly in the high moisture range. The high moisture bales which had been treated with propionic acid at the '2 per cent' level, received a calculated 2.5 per cent on average, but at all the other treatment levels the intended rates of application were achieved with reasonable accuracy.

Main Effects

TABLE 13

	High moisture		Low moisture					L.S.D.
Initial Moisture Content %	36.9	34.8	27.3	25.8	24.5	24.3	21.3	
Propionic acid % by wt.	2.5	4.9	nil	2.1	3.2	5.0	nil	
Initial W.S.C. % in D.M.	21.2	21.0	18.7	16.7	17.5	17.9	16.3	
Final W.C.C. % in D.M.	14.3	14.1	9.0	16.7	17.4	18.2	15.6	6.2
Loss of W.S.C. %	36.7	37.4	55.7	4.4	3.9	+2.6	5.2	44
Final Moisture Content %	25.1	25.4	16.2	20.7	22.5	21.4	15.4	NS
Moisture Loss %	43.9	39.5	52.5	28.2	13.1	14.8	33.2	
Dry Matter Loss %	6.1	5.4	7.4	4.2	3.1	0	1.0	NS
Degree days above ambient temperature	656	478	919	400	282	231	-	437

P = 0.05

TABLE 14

INDIVIDUAL BATCH RESULTS IN THE SECOND BATCH EXPERIMENT

Batch	Initial % H_2O	Block	% acid level	Initial W.S.C.	Final W.S.C.	% Loss W.S.C.	Final % H_2O	% H_2O Loss	% DM Loss	°Days above ambient
1	39.5	A	2.6	22.3	16.2	31.1	27.2	45.8	5.1	586
2	38.2	B	2.7	19.7	13.3	36.0	24.8	49.5	5.2	742
3	32.7	C	2.4	21.6	13.4	42.9	25.1	36.4	7.9	639
4	32.5	B	5.3	21.6	15.2	34.9	24.3	37.9	7.5	399
5	36.9	C	4.5	20.1	10.1	50.4	25.1	43.5	1.2	670
6	34.9	A	4.8	21.4	16.9	26.9	26.7	37.2	7.4	365
7	28.3	A	nil	19.1	10.9	48.0	16.5	54.4	8.9	1057
8	26.2	B	nil	18.2	7.1	63.3	15.8	50.7	5.9	782

TABLE 14 ctd.

Batch	Initial % H_2O	Block	% acid level	Initial W.S.C.	Final W.S.C.	% Loss W.S.C.	Final % H_2O	% H_2O Loss	% DM Loss	°Days above ambient
9	24.4	B	1.9	16.5	18.3	+3.6	18.9	32.5	6.6	206
10	27.8	C	2.3	15.8	15.7	+2.4	21.5	26.7	+3.0	623
11	25.2	A	2.1	18.0	16.0	19.1	21.7	25.3	9.0	370
12	24.8	B	3.1	17.4	15.8	9.4	22.8	10.5	0.2	302
13	23.1	C	3.3	16.6	17.5	0.1	22.8	6.8	5.2	211
14	25.7	A	3.2	18.6	18.9	2.2	21.9	21.9	3.8	331
15	24.8	A	5.0	17.5	21.1	+21.9	21.3	17.0	+1.1	253
16	24.0	C	4.9	18.8	16.5	14.9	22.5	10.9	3.0	237
17	24.0	B	5.0	17.3	17.1	+0.9	20.5	16.4	+2.0	206
18	21.3	barn dried	nil	16.3	15.6	5.2	15.4	33.2	1.0	-

The results for the untreated control batches indicated that a considerable degree of moulding and deterioration had taken place. More than half of the soluble carbohydrates were lost and almost twice as much heating took place as in any of the other treatments. As a result of the heating the moisture content fell from 27 per cent to 16 per cent, representing a moisture loss of over 50 per cent. The dry matter losses were not so high, however, at 7.4 per cent and were not in fact significantly above zero.

In the low moisture hays which received the propionic acid treatments of 2, 3 and 5 per cent, less moulding and deterioration appears to have taken place. Heating in the batches at all three acid levels was significantly below that in the untreated batches and the loss of soluble carbohydrates was reduced to a level not significantly above zero. Although the results for the loss of dry matter and moisture in these batches were below those in the untreated batches the differences were not significant.

The increasing levels of propionic acid applied to the low moisture hay brought very little if any reduction of losses, and none of the differences between batches with the different acid levels were significant.

In the high moisture treated hays losses were much higher than in the low moisture treated hays and were not significantly below those in the untreated control batches. The lower losses of dry matter, soluble carbohydrate and moisture in the high moisture treated batches when compared with the control batches, although not statistically significant, should be noted in view of the fact that the moisture contents of the control batches were on average 7 or 8 per

cent below those of the treated batches. The extent of heating the high moisture treated batches also fits the above pattern, not being significantly lower than the heating in the control batches.

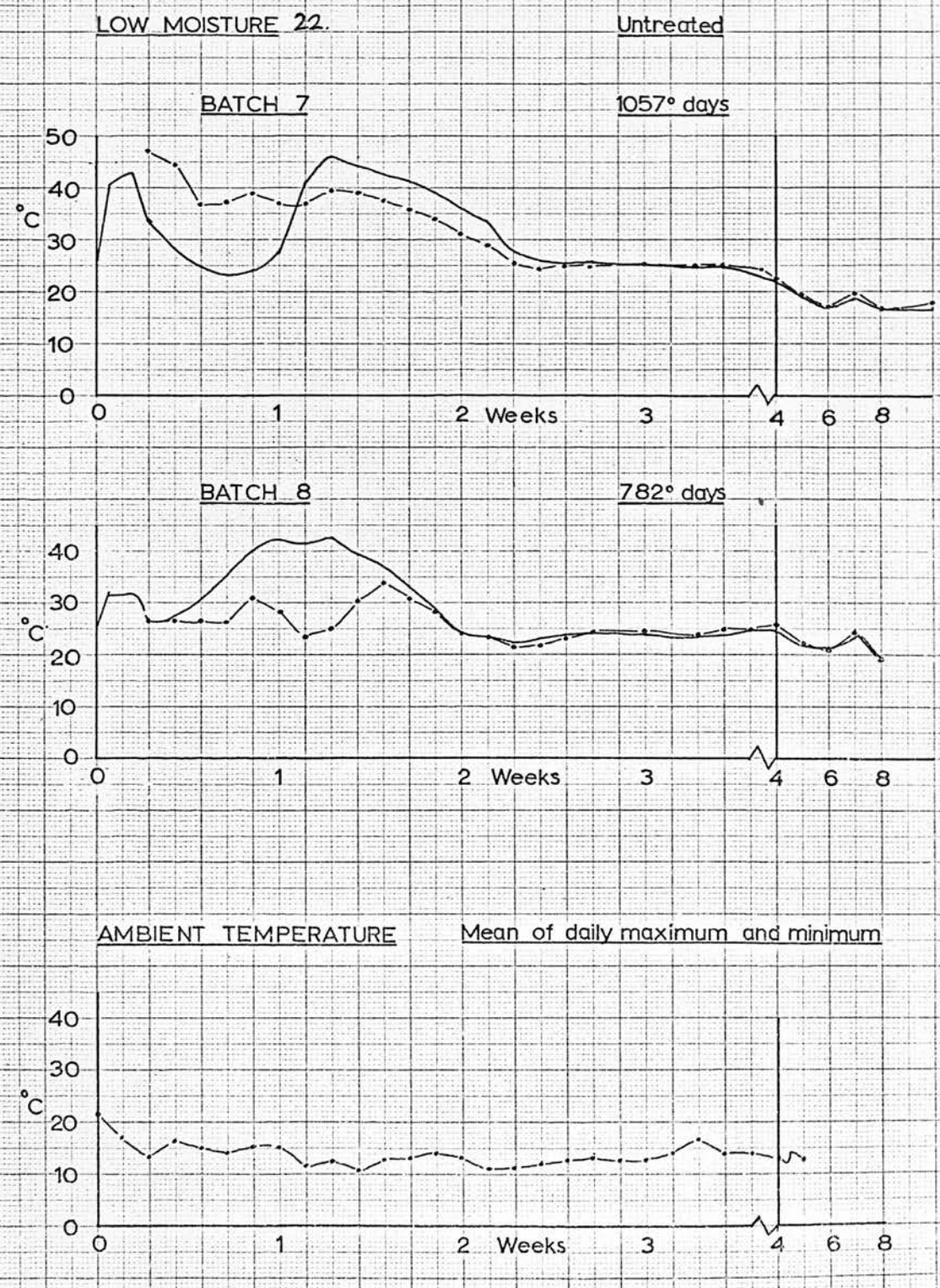
Losses in the barn dried hay were small and very similar to those in the acid treated low moisture hays.

The Pattern of Temperature Changes Figures 22 to 24

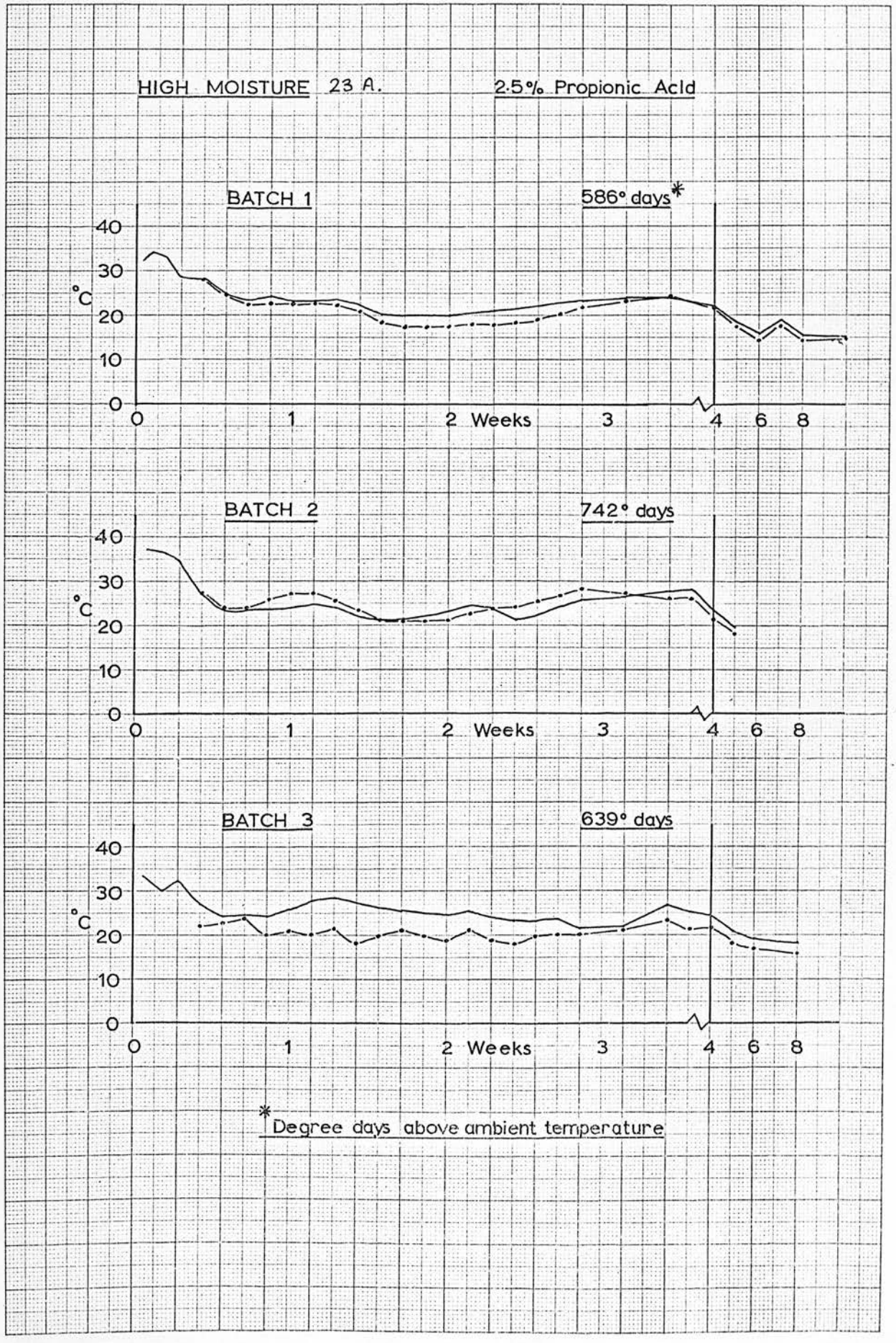
The temperatures in the untreated batches (Figure 22) followed the expected pattern with an initial peak in the first two days followed by a secondary peak at the end of the first week with a maximum in the region of 45°C.

The temperatures declined to the ambient by the end of 5 to 6 weeks. A measure of the extent of heating is given by the number of 'degree days above ambient'. These results have already been mentioned in the previous section and were obtained for each batch by accumulating the total over the first 5 weeks of the number of degrees Celcius above ambient each day. In the two untreated batches the totals were 1057 and 782 degree days.

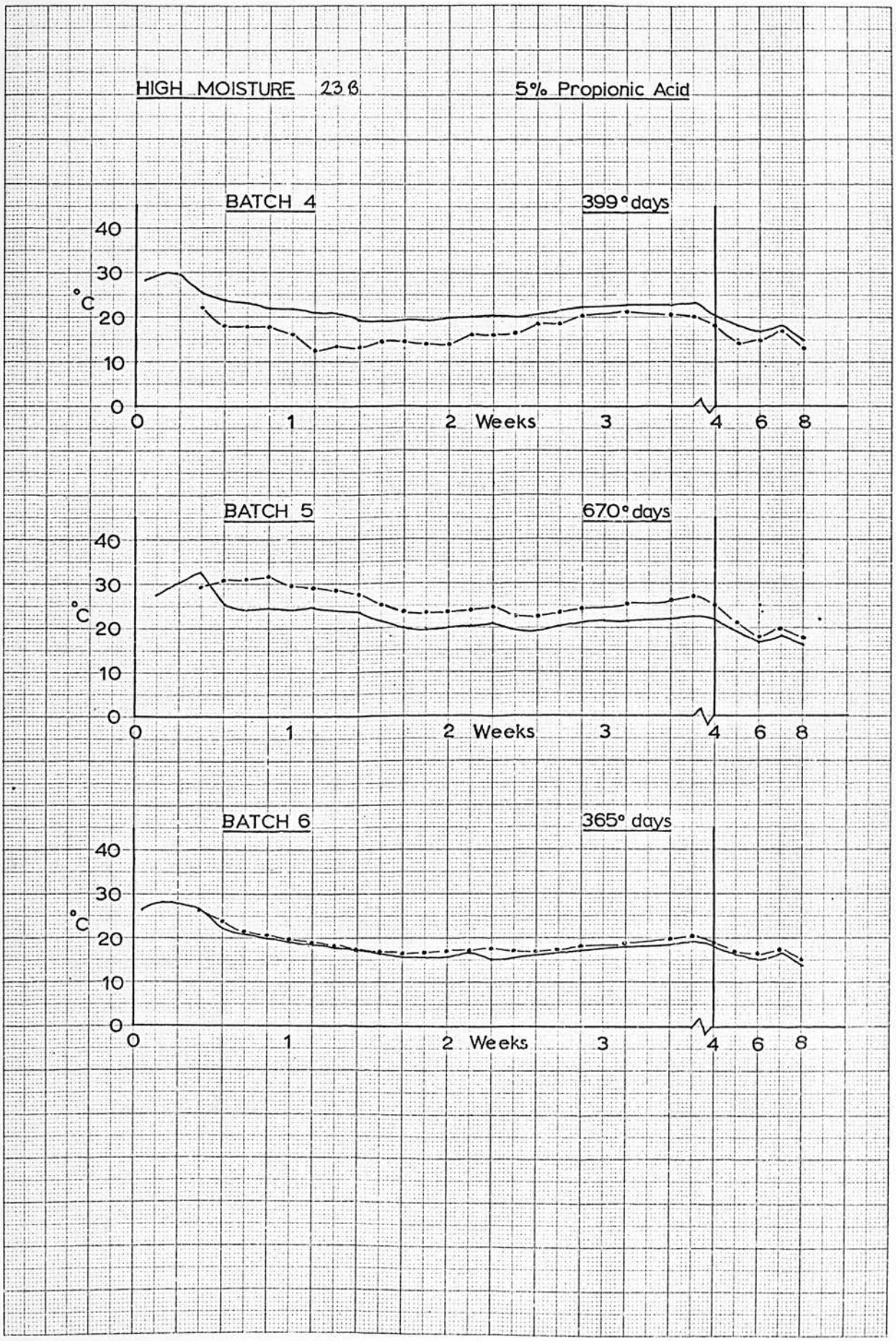
In all the propionic acid treated batches the temperature rose to an initial peak of between 30° and 40°C during the first two or three days (Figures 23 to 24). In the high moisture batches, Figure 23, these peaks were followed by temperatures consistently above the ambient temperature, particularly at the 2.5 per cent acid level. The batches with the 5 per cent acid treatment tended to have slightly lower temperatures as may be seen by comparing the degree day results. Batches 4 and 6 had the lowest numbers of degree days, and from table 14 it will also be seen that these batches received the highest acid

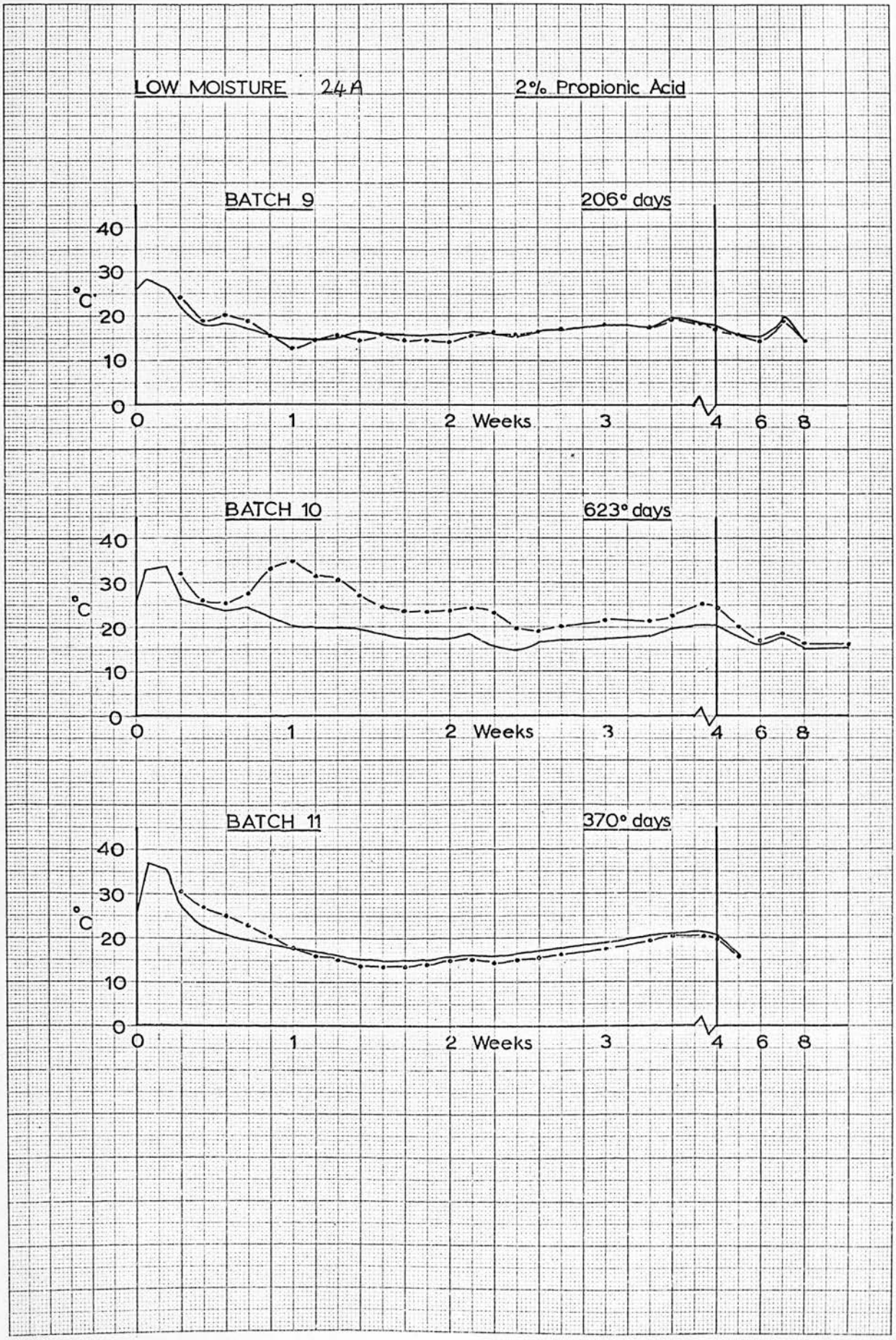


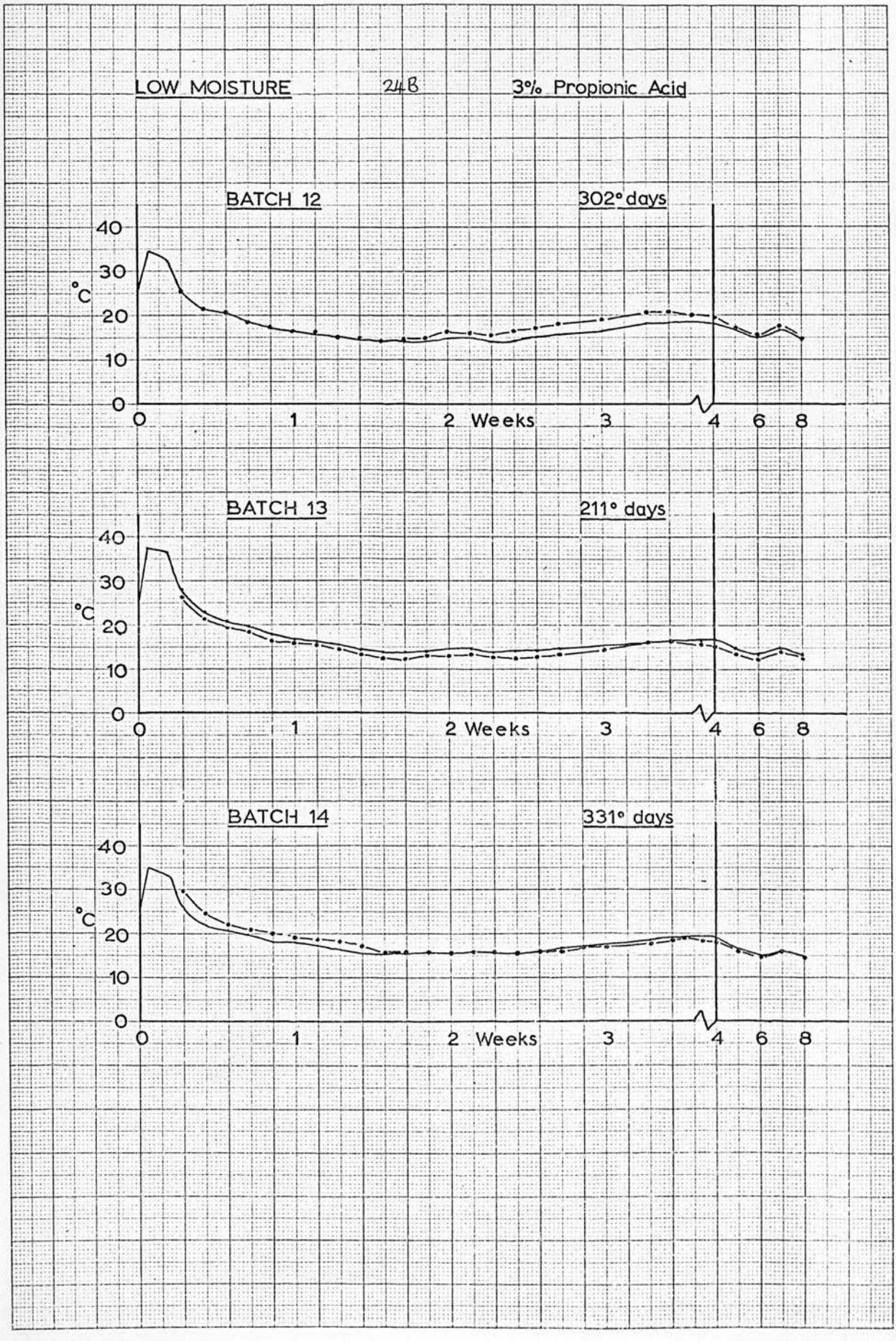
Each line represents readings from one thermometer.

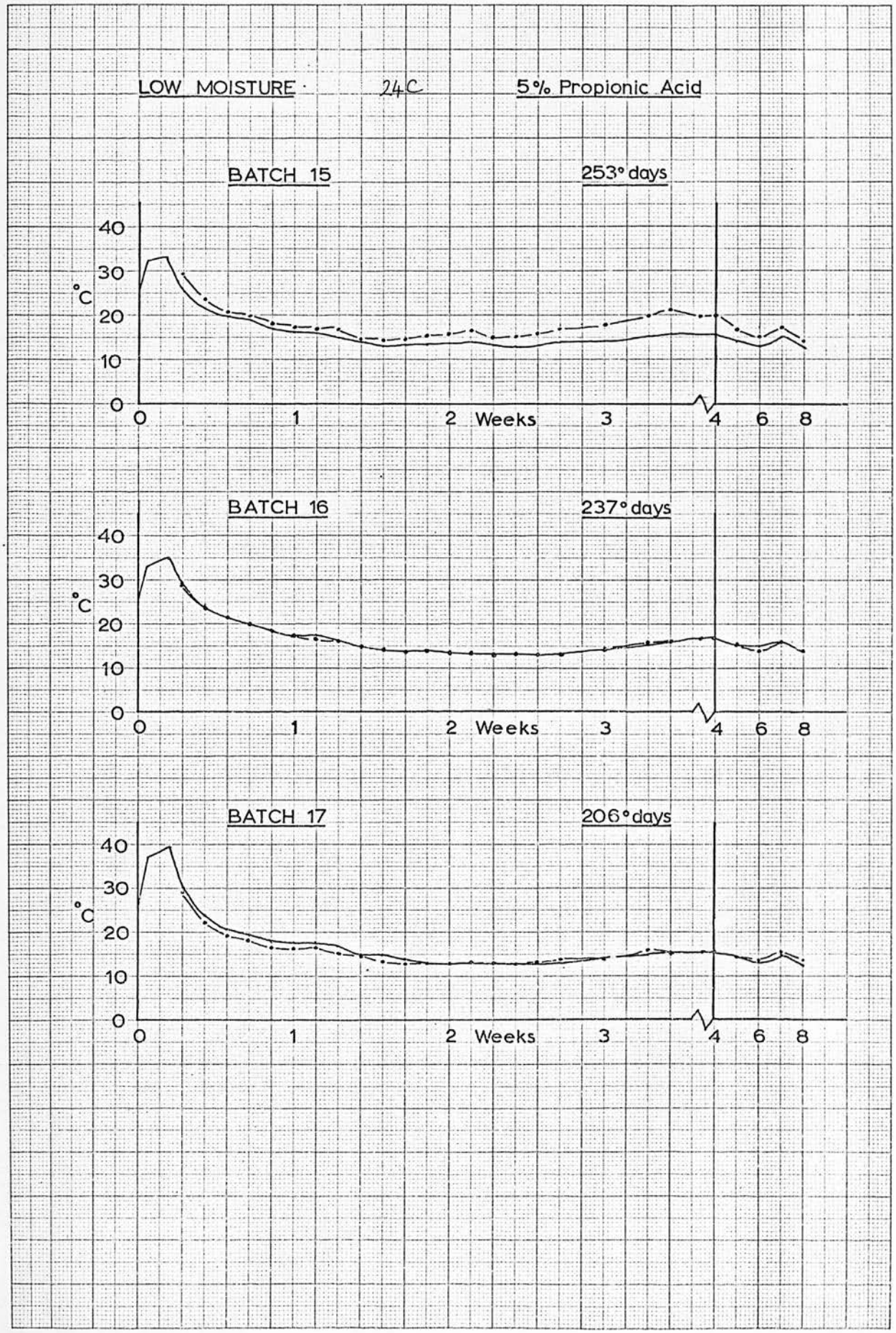


*Degree days above ambient temperature


HIGH MOISTURE 23%
5% Propionic Acid
BATCH 4
399° days
BATCH 5
670° days
BATCH 6
365° days
°C
40
30
20
10
0
0
1
2 Weeks
3
4
6
8


LOW MOISTURE 24A
2% Propionic Acid
BATCH 9
206° days
BATCH 10
623° days
BATCH 11
370° days
°C
40
30
20
10
0
0
1
2 Weeks
3
4
6
8


LOW MOISTURE
24B
3% Propionic Acid
BATCH 12
302° days
°C
40
30
20
10
0
0
1
2 Weeks
3
4
6
8
BATCH 13
211° days
°C
40
30
20
10
0
0
1
2 Weeks
3
4
6
8
BATCH 14
331° days
°C
40
30
20
10
0
0
1
2 Weeks
3
4
6
8


LOW MOISTURE
24C
5% Propionic Acid
BATCH 15
253° days
BATCH 16
237° days
BATCH 17
206° days
°C
40
30
20
10
0
0
1
2 Weeks
3
4
6
8

treatment of the high moisture hays, 5.3 and 4.8 per cent respectively, and also had relatively low moisture contents at 32.5 and 34.9 per cent respectively.

After the initial peak the temperatures in the low moisture treated batches declined almost to the ambient, the lowest temperatures being recorded in the batches which received the 5 per cent acid treatment (Figure 24c). Only in batch 10 was their any evidence of secondary heating such as might be expected if moulding was taking place. Of the low moisture batches, this batch had the highest moisture content at 27.8 per cent and received an acid treatment of 2.3 per cent (Figure 24a).

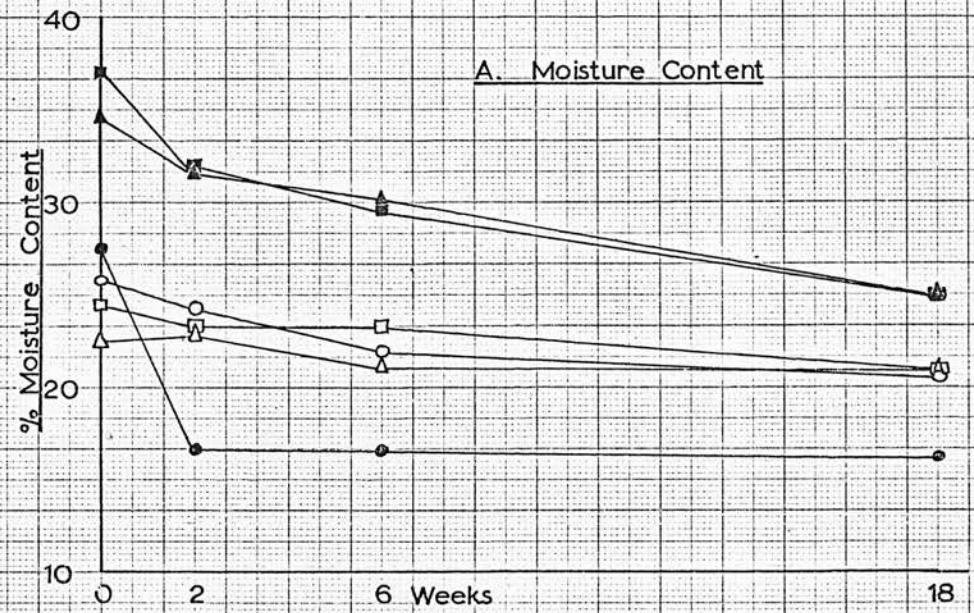
Pattern of Chemical Changes during Storage

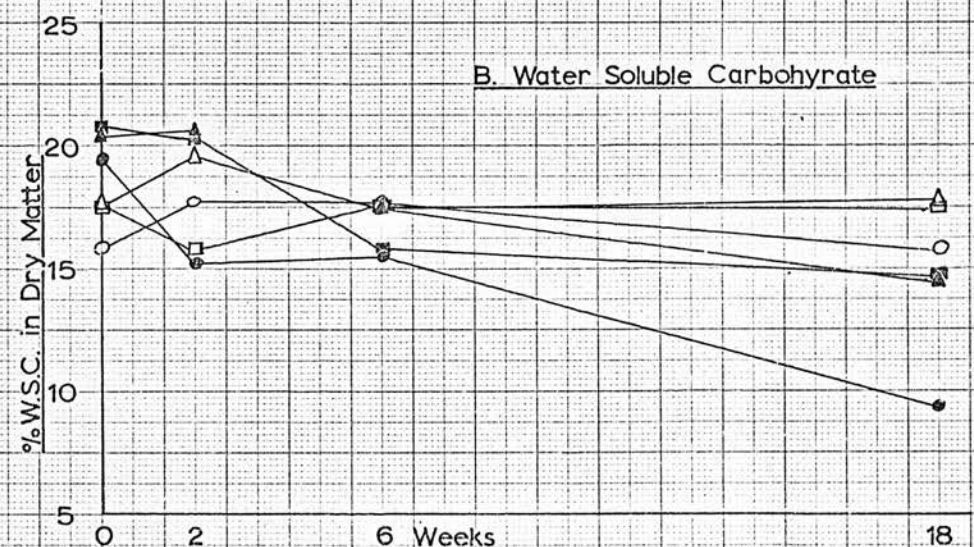
The results of the 2 week and 6 week samplings are shown along with the initial and final values for the moisture content and water soluble carbohydrate contents of the hays in Figure 26.

It will be seen that in the untreated hay virtually all the loss of moisture took place during the first two weeks of storage, corresponding with highest temperatures in these stacks. In the high moisture treated hays loss of moisture took place most rapidly during the first two weeks, but continued at an only slightly reduced pace during the 2 to 6 week and the 6 week to 18 week periods. This again may be correlated with the fact that in these batches heating, although reaching a peak in the first two weeks, continued well above ambient for most of the storage period.

The moisture contents of the low moisture treated batches changed very little during the 18 week storage period, dropping at the most by a few per cent, and all average out at about 21 per cent moisture,

FIGURE 26 CHANGES DURING STORAGE





High Moisture		Low Moisture			
■	2.5% acid	●	Untreated	□	3% acid
▲	5% acid	○	2% acid	Δ	5% acid

irrespective of the acid treatment applied. In the same way both the 2.5 and 5 per cent treated high moisture hays finished up with about 25 per cent moisture.

Changes in the water soluble carbohydrate content of the low moisture treated hays were negligible. Only the control low moisture hay showed a sharp decline in this fraction of the dry matter during the first two weeks of storage, again corresponding with the maximum heating in these batches. The decline in water soluble carbohydrate continued, however, in the untreated batches during the 6 to 18 week period.

The water soluble carbohydrate content of the high moisture untreated batches also showed a tendency to decline, but at a fairly steady rate, at both the 2.5 and 5 per cent acid levels, and followed a similar pattern to the moisture changes.

The pattern of Microbiological Changes Table 14

The microbiological counts from the samples taken at 2 weeks and 6 weeks show the untreated hays to have significantly higher mould counts than any of the treated hays. In these control batches at the end of 2 weeks during which considerable heating had taken place, reaching temperatures in excess of 40°C, similar counts of moulds on the plates at the 27° and 36°C incubation temperatures were obtained. At the end of six weeks of storage, however, the count of moulds growing at 27°C had increased whereas that at 36°C had fallen. These changes may be linked with the temperature in the batches, which had fallen also to about 25°C.

The variations in the mould counts in the acid treated hays are

too great to allow reliable conclusions to be drawn from them (See full results in appendix 2), but from table 14a it will be seen that they are generally highest in the 2 week sample at 27°C. The slight rise in the mould counts at 27° and 36°C in the low moisture batches treated with 2 per cent acid, against the opposite trend seen at the higher acid levels may be linked with the secondary heating which took place in batch 10.

TABLE 14

SECOND BATCH EXPERIMENT

Mean Microbiological Counts at each Treatment Level

MOULD COUNTS

		27°C		36°C	
Moisture Level	Acid %	2 week	6 week	2 week	6 week
High	2.5	18	41	10	13
High	5	14	23	6	5
Low	nil	324	475	322	248
Low	2	31	75	14	32
Low	3	44	6	14	3
Low	5	46	5	16	2
Barn Dried	nil	40	73	14	9

L.S.D. P = 0.05 256

TABLE 14 ctd.

BACTERIAL AND ACTINOMYCETE COUNTS

		27°C		36°C		55°C	
Moisture Level	Acid %	2 week	6 week	2 week	6 week	2 week	6 week
High	2.5	17	5	10	5	85	2
High	5	13	116	17	129	3	4
Low	nil	105	129	14	7	2	2
Low	2	16	22	11	2	1	3
Low	3	9	4	8	3	0	3
Low	5	5	5	9	3	1	2
Barn Dried	nil	56	20	148	6	199	2

The untreated control hay also had the highest counts of bacteria and actinomycetes at the 27°C incubation temperature. The counts of these organisms were low, however, at 36°C and this may suggest that the heating which took place was a result of fungal activity and not of bacteria and actinomycetes. None of the low moisture propionic acid treated hays had high counts of these organisms, although it is again of interest that the 2 per cent treated bales did have slightly higher counts than the batches at the higher acid levels. None of these batches had high counts of thermophilic actinomycetes at 2 weeks or 6 weeks.

One of the samples from the high moisture treated batches had a high count of bacteria and actinomycetes at both 27°C and 36°C at 6 weeks. It will be seen from the full results in appendix 2 that this sample originated from batch 4 which had in fact received the highest

level of acid, 5.3 per cent, and had the lowest moisture content, 32.5 per cent, of the high moisture batches. One sample from batch 2 at 2 weeks had a high count of thermophilic actinomycetes, but this result was not confirmed in the replicate plate made from the same sample.

The barn dried batch of hay had relatively high counts of bacteria and actinomycetes at all three incubation temperatures in the 2 week sample, but the 6 week sample was almost free from such contamination.

Assessment of Visible Moulding at the End of Storage Figure 27

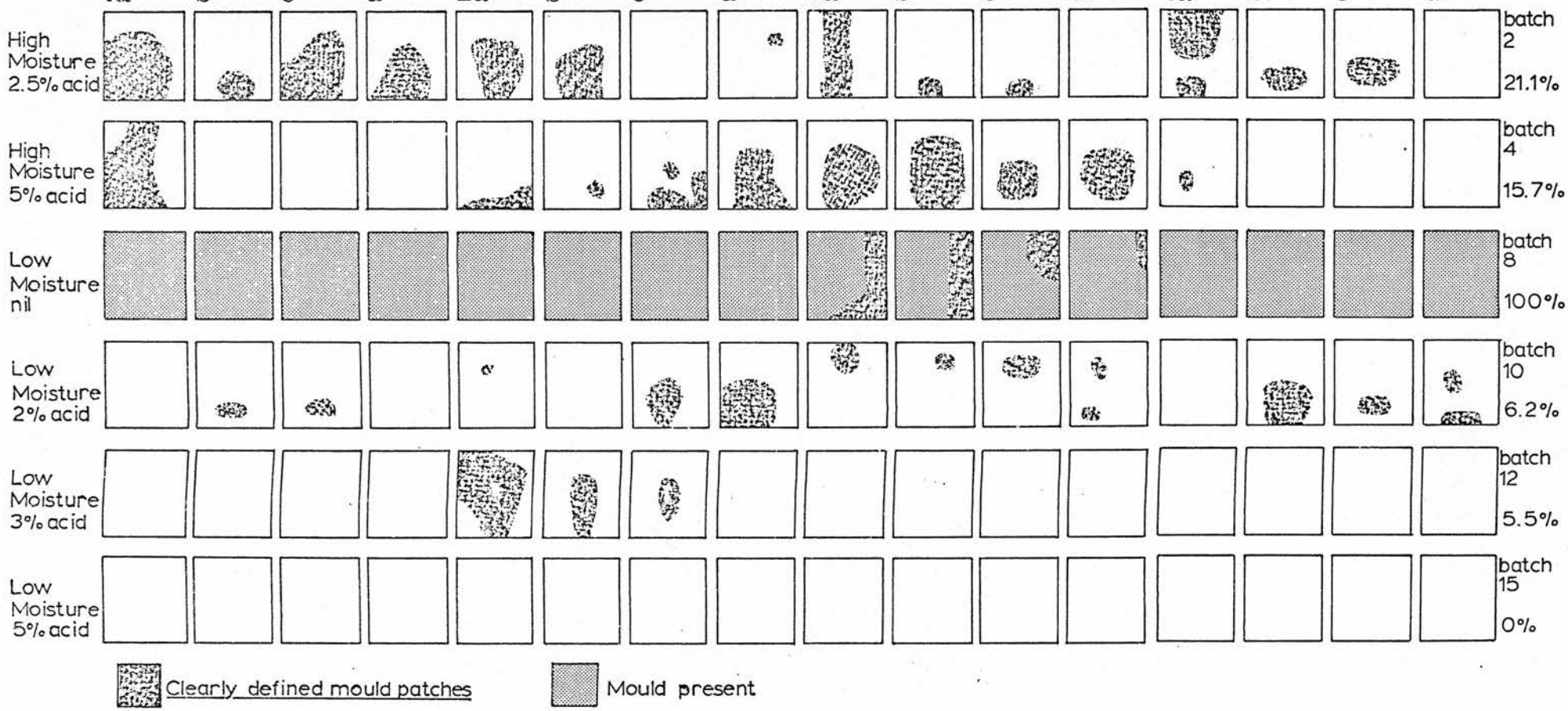
The figure depicts diagramatically the moulding as seen in the parts of the four bales from each of the 6 batches indicated at the right hand side. The figure below the batch number is the estimated per centage of mouldy hay in the 4 bales examined.

The untreated control bales were 100 per cent mouldy although only in one of the bales could clearly defined patches of mould be seen. In the others the moulding could be detected by a musty smell and by the dry and dusty appearance of the hay.

In the high moisture propionic acid treated hays patches of mould growth were found in all the bales, and extensive moulding in a number of the bales. The well preserved parts of the bales were an olive green colour whereas the hay in the mouldy parts usually became a darker colour with the growth of white or yellow moulding evident. Mould growth in the high moisture hay which received 5 per cent propionic acid was not significantly less than in the hay with 2.5 per cent acid.

Again in the low moisture bales from the 2 per cent acid treatment patches of mould growth were found in all the four bales examined. The moulding, however, was much less extensive than in the high moisture

FIGURE 27 PATTERN OF VISIBLE MOULDING Four randomly selected bales from one batch at each treatment level



treated batches, amounting to about 6 per cent of the bales. The bales at this treatment level were taken from batch 10 which has already been mentioned as showing more heating and having high mould counts than the other low moisture treated batches.

Only one of the bales from batch 12, which received the 3 per cent acid level, contained mould patches, although the moulding in this one bale was quite extensive. None of the bales from batch 15, which received the 5 per cent acid treatment contained any visible moulding.

TABLE 16

Comparison of the Results of the First and Second Batch Experiments

First Batch Experiment — 9-bale batches

Initial Moisture Content %	Acid Level %	Dry Matter Loss %	W.S.C. Loss %	Moisture Loss %	Mould %	Max. Temp °C
34	2.3	12.4	56.5	40.4	35	29
40	4.1	2.6	40.1	36.9	20	29
27	nil	7.4	56.9	53.7	100	45
27	2.4	4.7	+3.1	4.2	0	20
28	3.4	1.0	+9.0	13.9	0	21
26	4.5	3.6	8.3	5.3	0	20

TABLE 16 ctd.

Second Batch Experiment 50-bale Batches

Initial Moisture Content %	Acid Level %	Dry Matter Loss %	W.S.C. Loss %	Moisture Loss %	Mould %	Max. Temp °C
37	2.5	6.1	35.6	45.1	21	38
35	4.9	7.1	37.2	39.5	16	33
27	nil	7.1	55.7	52.5	100	47
26	2.2	2.7	4.4	28.1	6	37
25	3.2	0.8	6.6	16.8	5	38
24	5.0	0	+2.6	14.8	0	39

DISCUSSION

As the aim of this experiment was to investigate the validity of the conclusions of the first batch experiment with regard to relatively large batches of hay as well as small 9 bale batches, the results of this experiment are best seen in comparison with those of the first batch experiment. Table 16 brings together the relevant results from both experiments.

The corresponding untreated batches behaved very similarly with losses of dry matter, moisture and water soluble carbohydrate which were identical or almost so in each case. The temperature patterns too, which may be seen in figures 12 & 21 are very similar with maximum temperatures of about 45°C being reached during the second week of storage. This result would tend to confirm the conclusion of Gregory et al, (1963) that with regard to untreated hay the changes taking place in 9 bale stacks do correspond favourably with those taking place in larger stacks of bales.

In the propionic acid treated hays also many similarities can also be seen in the results of the two experiments, although the correlation is not as close as in the untreated hay. The slight differences in the initial moisture contents and acid treatment levels between the two experiments are bound to have been partly responsible for this variation. The 50 bale batches tended to reach a higher temperature than the 9 bale batches, but from the graphs it will be seen that all of these maximums were during the first 2 or 3 days of storage. During the rest of the storage period the temperatures were very similar, and if anything, slightly lower in the 50 bale batches. The ambient temperature at the time of baling was higher in the second experiment and this may have been a cause of

the higher initial peaks in the 50 bale batches.

In the high moisture treated hays moulding appears to have been more extensive in the 9 bale batches and this may be linked with the slightly lower acid levels applied to these hays. Water soluble carbohydrate losses followed the same trend although the other losses were not so consistent. The low moisture acid treated batches in both experiments had low losses of dry matter and water soluble carbohydrate, but moisture losses tended to be higher in the 50 bale batches. The mouldy patches amounting to 5 or 6 per cent of the hay at the 2 and 3 per cent acid levels in the 50 bale batches were not found in the corresponding 9 bale batches. The highest acid levels in both experiments, however, entirely prevented visible moulding.

Overall, therefore, it may be concluded that the 50 bale batches in this experiment did not produce results materially different from those found previously in the 9 bale batches.

While the experiment may have been successful in achieving its main aim, some mention should be made of the problems encountered. A glance at the full results in table 14 reveals how wide the variations were between batches which were meant to be identical replicates with the same moisture content and acid treatment. It is not surprising therefore that the final results showed wide divergences and that it was difficult to find statistically significant differences. Many factors could have contributed towards this variation and some of the more important ones are not difficult to find. The condition of the hay in the field at the time of baling, for example, is likely to have been an important factor.

It has already been pointed out in the results section that due

to the fine weather conditions at the time of baling the hay was drying out very rapidly. The baling of the 50 bales for any one batch took between 10 minutes and 15 minutes so that, allowing time for recalibration of the applicator between batches the three replicate batches at one treatment level would take between 45 and 60 minutes to complete. The rapid drying of the hay would therefore give rise to a trend for batches to have lower and lower moisture contents as time went on.

Another problem encountered in the field as a consequence of requiring 50 bales for each batch was the difference between the swaths. Ideally, in a field with an entirely uniform sward at cutting and which is tedded and turned and windrowed properly, the final swaths should be all very much the same in the quantity of hay they contain, the moisture content of that hay and in the uniformity of the hay along the swath. These conditions, however, rarely occur and certainly did not in this experiment. Whether due to variations in the original sward or to uneven tedding, turning or windrowing, the swaths were clearly different in the quantity of hay they contained. The differences were not such as would normally bother a farmer making hay, but were probably of importance in this experiment for two reasons.

Firstly, hay will tend to dry more quickly in a thin swath than a thick one, and therefore swaths uneven in thickness will also become uneven in moisture content. In this experiment any unevenness in moisture content between swaths would result in an additional variation within the batches over and above that already likely to be present due to minor variations in moisture content within each swath. In the distribution experiments which are described in section three of the thesis the relationship of moisture content variations together with propionic acid distribution to the occurrence of mould patches is investigated,

but even without consulting these experiments, it is obvious from this experiment that moisture content variation and the occurrence of mouldy patches are highly likely to be related.

As well as giving rise to moisture content variations, the differences between swaths is also likely to have affected the rates of propionic acid reaching the hay. Where the acid is applied at a constant rate and the baler is moving forward at a constant speed the percentage level of acid being applied to the hay will vary inversely in proportion to the quantity of hay per unit length of swath. (ie. more hay means a lower acid treatment and vice versa.) As mentioned earlier, each batch required the baling of two or three complete swaths, and during this baling the application rate and tractor speed were not altered. Thus as a result of the variation between the swaths it is likely that each batch contained bales of quite widely differing acid treatment. The variation in acid treatment is therefore another factor which is likely to have contributed to the differences between replicates and also to the irregular distribution of mouldy patches within and between bales of each batch (Figure 27).

While these variations have been considered here in the light of a scientific investigation, they also have a bearing on the practical use of propionic acid by farmers as a hay preservative. Variations between swaths are likely to be greater on a commercial farm in which particular care is not being taken in this direction, and the problems of uneven moisture content and acid treatment are going to be intensified when the farmer is baling not 50 but 2000 bales in a batch.

Also of practical importance in the prospective use of propionic acid commercially is the ease of operation of the applicator. The low

pressure applicator used in this second experiment was more easily fitted to the baler than the high pressure applicator of the first experiment, could be calibrated more easily, had a wider range of application rates and created less problems during its operation. The most difficult problem associated with its use was priming the pump, which had to be carried out each time the 5 gallon acid 'flo-pack' was replaced (see Appendix 7). Whether this applicator was any more efficient at distributing the acid evenly through the bales is not possible to say, but its use did not prevent the occurrence of mould patches as in the first batch experiment. In the distribution experiment in section three, however, the effectiveness of the low pressure applicator is investigated.

In this experiment one of the treatments, namely the 5 per cent propionic acid treatment of the low moisture hay with an average moisture content of 24 per cent, reduced losses virtually to zero and prevented the occurrence of visible moulding. A 5 per cent treatment level is, however, more than twice the level which might be considered as economically feasible, and a moisture content of 24 per cent is almost below the range in which a farmer might consider a preservative treatment to be necessary. The three per cent acid treatment at this moisture level was almost as effective as the 5 per cent level at reducing losses but one of the four bales examined from this treatment level contained moulding. At the two per cent acid level one of the batches, the one with the highest initial moisture content showed definite signs of heating, contained widespread but not extensive moulding, the evidence being supported by increasing mould counts in the 2 to 6 week storage period. Dry matter and other losses were not significantly higher, but the indication is still clear that the 2.3

per cent acid treatment applied to this batch at 27.8 per cent moisture was not quite sufficient to provide satisfactory preservation of the hay under the conditions of this experiment.

The barn dried batch also had low losses of a similar order to those in the acid treated low moisture hays. It should be noted, however, that with an initial moisture content as low as 21.3 per cent the losses would not have been great even without barn drying.

At the higher moisture levels between 32 and 40 per cent, neither the 2.5 or the 5 per cent acid levels were adequate to prevent serious losses and widespread moulding from taking place.

SECTION TWO

FIRST MINI-BALE EXPERIMENT

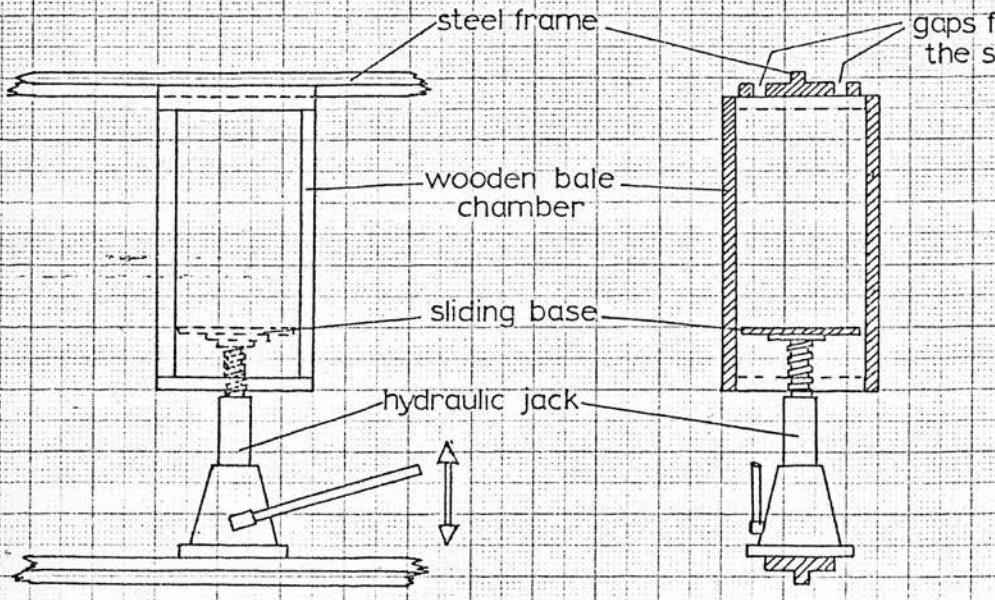
AIMS

The wide variations of moisture content and of propionic acid distribution in hay which was baled and treated with the acid in the field rendered the results of the batch experiments of little value for establishing the exact limits of the preserving effect of propionic acid on hay. Only by using an experimental technique which eliminates these variations would it be possible to obtain an accurate assessment of the levels of propionic acid application needed to preserve hay of different moisture contents. The need therefore was for a small scale experiment in which the hay could be mixed thoroughly to avoid moisture content variations and the acid applied in such a way as to ensure very even distribution. The conditions of the experiment would however have to resemble as far as possible those pertaining to conventional sized bales.

The use of small scale bales, or 'mini-bales' was thought to fulfil the above requirements best, small enough to allow easy mixing of the hay and accurate acid treatment and large enough to permit compression of the hay to densities similar to those of conventional sized bales. With a bale size of about 20 cm^3 and using hay of about 30 per cent moisture such bales would weigh about 1 kg. An experimental rig was prepared using a wooden 'chamber' and a hydraulic jack to compress the hay. Drawings of this rig and also of the Mark II Mini-baler used in the second experiment are shown in Figures 28A&B

The aim of this first mini-bale experiment was to investigate the properties of small bales and their value, if any, for the type of experimental work outlined above.

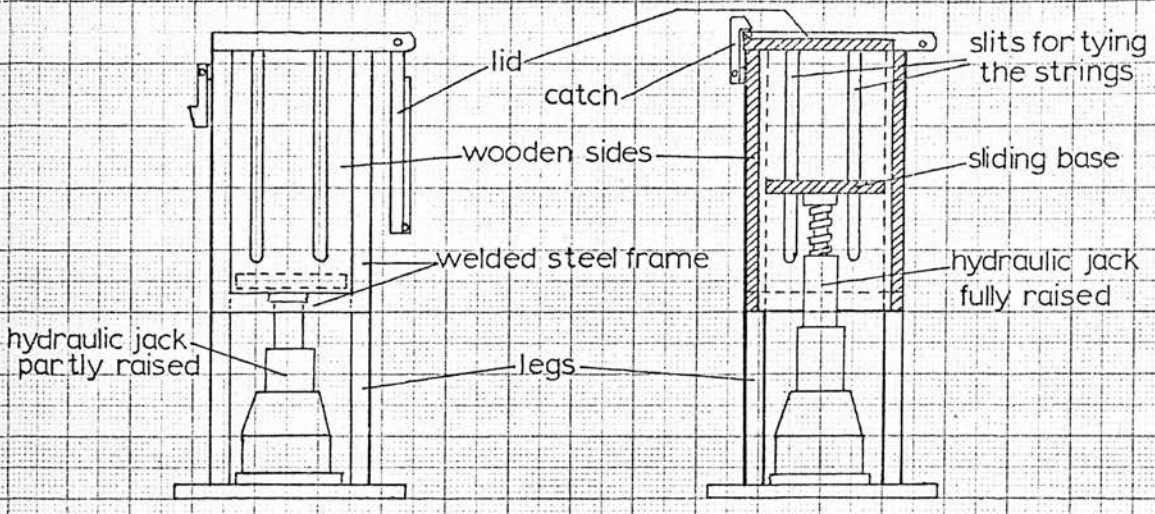
FIGURE 28A THE MARK I MINI-BALER



Elevation

Sectional End Elevation

FIGURE 28B THE MARK II MINI-BALER



Elevation with lid open

Sectional Elevation with lid closed

DESIGN

A simple experimental design was chosen which fulfilled the above requirements. The mini-bales were to be made from hay at one moisture content only, but treated with a number of levels of propionic acid. Obviously the choice of moisture level was critical and it was decided to aim for a relatively high moisture content in the region of 45 per cent which would be near the upper limit of moisture levels relevant in practical haymaking and yet within the range of moisture levels at which it was believed propionic acid could be effective.

The earlier experimental work had given no real indication of how much acid would be needed to preserve hay at this moisture level and so it was decided to use a wide range of propionic acid levels, 1, 2, 4, 6, and 8 per cent, in the hope that we would be including both adequate and inadequate levels. The design included four replicate bales at each of the acid treatment levels and during the storage period these bales were to be grouped in four randomised blocks each containing one bale of each treatment level.

METHODS

Field and Storage Work

A small area of pure perennial ryegrass was cut at the flowering stage and tedded both by machine and by hand to ensure rapid and even drying. However, the weather during this period was not ideal, being cool with showers and only occasional bright periods, and by the third day the moisture had only fallen to about 50 per cent. Although this was slightly above the level originally intended it was decided to go

ahead with baling as further drying in the field seemed unlikely under the prevailing weather conditions. Sufficient hay for the experiment was loosely filled into large polythene bags and taken to the store.

After being mixed thoroughly the hay was divided into six equal weighed lots in readiness for the acid treatments. Each lot of hay in turn was spread evenly over the concrete floor and the appropriate quantity acid applied evenly onto the hay using a hand sprayer. During spraying, the hay was turned two or three times to ensure even application. The sprayer was weighed before and after each treatment had been applied in order to establish the exact quantity of acid used.

On the completion of each treatment the hay was again thoroughly mixed and then divided into 4 equal portions by weight. Each portion was made into a mini-bale. The method of producing a mini-bale is given in Appendix 9.

The bales were stored in insulated boxes made of expanded polystyrene which were themselves kept in the insulated bins of a potato store. In this way fluctuations in the environment of the bales were kept to a minimum. A mercury thermometer was inserted through a hole in the lid of each box into the centre of each bale. The bales were stored over a 14 week period.

Sampling and Analysis

The moisture content of the hay as it dried in the field was followed using the same methods as outlined for the first batch experiment. After the hay had been brought in and mixed a number of grab samples were taken and mixed to provide material for a triplicate water soluble carbohydrate determination. Moisture content and pH

determinations were taken on grab samples taken from each lot of hay after the acid treatment had been supplied.

Immediately prior to storage each bale was trimmed to remove any straggling strands of hay which might have fallen off and caused errors, and then weighed to the nearest gram. The temperatures of the bales were recorded daily during most of the experiment, but towards the end of the three month storage period when little or no change was taking place the frequency of reading was reduced.

At the end of storage each bale was weighed and its visual appearance noted. Using the corer designed by Alexander et al (1969), 3 or 4 cores were taken through each bale, amounting to 15 to 25% of the volume of the bale. This sample was mixed and subsampled for biochemical analysis. Duplicate determinations were carried out for oven dry matter, pH and water soluble carbohydrate.

No form of mould assessment other than visual appearance was carried out as it was considered that at this high moisture level any moulding would in fact be clearly visible.

RESULTS

It was immediately clear that considerable decomposition had taken place in all the bales which received the nil, 1 and 2 per cent acid treatments. In these bales the hay had turned black in colour and the strings tying the bales had rotted; mould growth was clearly evident. In contrast, the bales treated with 4, 6 and 8 per cent propionic acid did not show visible signs of deterioration, the hay having kept its original colour. No moulding could be seen.

All the results of temperatures and biochemical analyses followed the same pattern, with clear signs of deterioration of the nil, 1 and 2 per cent treatments and very little or no deterioration of the 4, 6 and 8 per cent treatments (Table 17).

TABLE 17

FIRST MINI-BALE EXPERIMENT

Acid Level	nil	1%	2%	4%	6%	8%	L.S.D. P = 0.01
Initial Moisture Content %	50.4	51.0	51.1	50.2	51.3	51.0	
Final Moisture Content %	21.0	23.3	23.9	43.4	43.1	44.0	
Initial W.S.C. in Dry Matter %	13.7	13.7	13.7	13.7	13.7	13.7	
Final W.S.C. in Dry Matter %	0.6	0.9	0.9	15.7	13.6	14.5	
Initial pH	6.50	5.50	4.97	4.59	4.40	4.31	
Final pH	8.19	8.14	8.29	4.90	4.73	4.72	
Dry Matter Loss %	30.1	31.3	25.7	3.4	9.0	4.4	9.7
Moisture loss %	81.2	78.3	75.6	25.1	27.3	22.1	7.3
W.S.C. Loss %	97.0	95.7	95.9	+10.2	10.0	+0.7	12.9
Degree days above ambient	240	207	164	33	16	27	35

Temperatures (Figure 29)

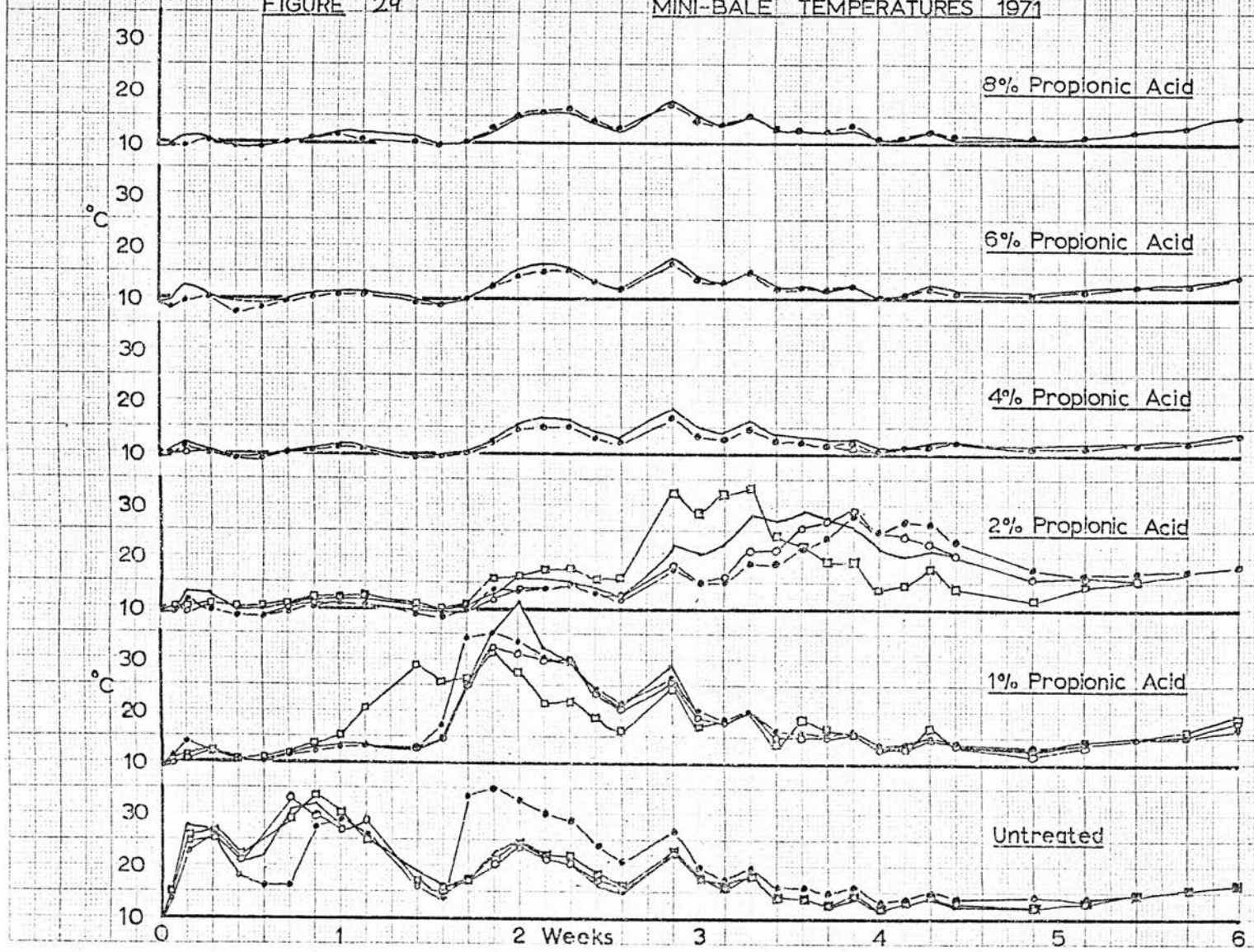
In the untreated bales rapid heating took place and an initial peak of 25° to 30°C was reached by day 2 or 3. This was followed by a slight drop in temperature and then a secondary, more substantial, peak of 30° to 35°C being reached at 5 to 6 days. Thereafter the temperatures of the bales declined to the ambient temperature after about 4 weeks. All four replicates followed a similar pattern, although one bale had temperatures considerably above the other three during most of the 2nd and 3rd weeks.

Heating was also evident in all the bales of the 1 and 2 per cent acid treatments but the onset of the temperature rise was delayed in each of these bales. From Figure 29 it is seen that there was no clearly defined initial peak, and that the main heating took place at the end of the 2nd week in the 1 per cent treatment, and during the fourth week in the 2 per cent treatment. The maximum temperatures reached by the 1 per cent treated bales were similar to those of the untreated hay, being in the 30° to 35°C range mostly, whereas the temperatures of the 2 per cent treated bales only reached between 25° and 30° in three out of the four bales.

In the bales treated with 4, 6 and 8 per cent acid the temperature pattern followed that of the ambient temperature throughout the 12 week storage period.

The overall extent of heating is indicated by the number of degree days above the ambient temperature. The highest total was found in the untreated hays with progressively lower totals in the 1 and 2 per cent treatments. In the mini-bales receiving 4 per cent acid and more very few day degrees above ambient were recorded. (Table 17)

FIGURE 29 MINI-BALE TEMPERATURES 1971



Losses

Very high losses of dry matter and water soluble carbohydrate took place in all the untreated, 1 and 2 per cent treated bales, reflecting the extent of heating and moulding in the bales (Table 17). No significant differences were found between these three treatments in terms of dry matter loss, water soluble carbohydrate loss, or moisture loss.

In the 4, 6 and 8 per cent treatments losses were in every case significantly lower than those in the 1, 2 per cent and untreated bales. Moisture losses amounted to 22 to 27 per cent of the original moisture, and dry matter losses were below 10 per cent. Differences between the three acid levels, however, were not significant. The water soluble carbohydrate losses were more variable, the 8 per cent treatment losing none, the 6 per cent treatment losing 10 per cent and the 4 per cent treatment gaining 10 per cent. The latter two results were significantly different from each other, and from zero, at the 1 per cent level of significance.

Accompanying the moulding in the bales of the 2, 1 and nil per cent treatment levels was a rise in the pH from about pH 5 in the acid treated and 6.5 in the untreated bales to between 8.1 and 8.3. In the twelve well preserved bales on the other hand the pH values rose only slightly, all remaining below pH 5.

DISCUSSION

Although a high moisture content was planned for the hay in this experiment, it was not intended to be as high as 50 per cent. However, the poor weather conditions left us with the choice of baling at this moisture content, or delaying the experiment for quite a number of days, no doubt with much loss in the quality of the hay. As it turned out, however, the hay of 50 per cent moisture satisfactorily demonstrated the ability of propionic acid to preserve high moisture hay.

It is clear from the results that the three high acid level treatments effectively preserved the mini-bales, suppressing mould development, preventing heating and reducing losses to a minimum. In a few of these bales dry matter losses over 10 per cent were found which would seem rather high in view of the otherwise excellent preservation. It may have been that surface losses by oxidation and physical loss during handling was relatively greater in these bales than would be expected in conventional sized bales due to the greater surface to volume ratio of the mini-bales. Of the three bales which had dry matter losses of over 10 per cent, two also had relatively high water soluble carbohydrate losses, 13 and 17 per cent. Although no visible moulding was present in these bales, it is possible that some biological deterioration, possibly due to bacteria or yeast took place.

In a number of bales apparent increases in water soluble carbohydrate were found. If, however, an average is taken of all the bales which were well preserved, ie. those of the 4, 6 and 8 per cent acid treatments, there was an overall net loss of 1.1 per cent. The initial value for the water soluble carbohydrate was derived from a single bulked sample and would not therefore indicate slight

differences between individual batches at the start of the experiment. Where such errors did arise then an apparent gain or loss of water soluble carbohydrate would be recorded. It may however have been that the increases in water soluble carbohydrate were real, although no satisfactory explanation can be given as to how they occurred. Similar apparent rises were found in the first batch experiment.

The results of this experiment are perhaps of most interest when compared with those of the First Batch experiment, as seen in Table 18.

The mini-bales which received the 2 per cent acid treatment were uniformly mouldy, and virtually complete loss of the water soluble carbohydrate took place. In the most nearly corresponding bales of the first batch experiment, at the high moisture level, moulding occurred in patches, and was found on 60 per cent of the hay. Water soluble carbohydrate, dry matter and moisture losses were lower in the full sized bales than in the mini-bales at this treatment level. The maximum temperatures recorded in the full sized bales were, however, higher than in the mini-bales. The mini-bales would, of course, tend to loose heat more rapidly from their proportionately larger surface area than the conventional sized bales, so that, in spite of the insulated boxes in which the mini-bales were kept, the temperature rises would be less even if the rate of biological activity was the same or greater.

At the 4 per cent level of acid application the picture is quite different. No moulding was present in the mini-bales and no loss of water soluble carbohydrate took place, while in the normal sized bales 20 to 35 per cent of the hay was mouldy and over 40 per cent of the water soluble carbohydrate was lost. This took place in spite of the

fact that the normal sized bales had moisture contents 10 per cent below those of the mini-bales. Moisture losses were also very much lower in the mini-bales treated with 4 per cent propionic acid, but the dry matter losses seem high in comparison with those in the medium moisture bales of the first batch experiment. Possible reasons for the relatively high dry matter losses in the mini-bales have already been discussed. The maximum temperature of the 4 per cent treated mini-bales indicates that no heating took place, whereas in the corresponding full sized bales heating up to a temperature of 44°C was recorded. Thus while the 4 per cent acid treatment was entirely effective in preserving the mini-bales, it was not nearly so effective when applied less evenly to hay with a more variable moisture content during the baling of conventional sized bales, even at a lower average moisture content.

The contrast between the results for the mini-bales and the conventional sized bales at these two acid levels also demonstrates how much more useful the mini-bales are in this type of experimental work. In the mini-bales the difference between the mouldy 2 per cent treated bales and the mould free 4 per cent treated bales is quite dramatic, whereas the differences between the 2 and 4 per cent treated conventional bales at either the high or medium moisture levels are much less clear-cut and in the analysis turn out to be not statistically significant in most cases. The value of the mini-bales as an experimental tool has therefore been adequately demonstrated.

In the mini-bales which deteriorated, once moulding had set in all the bales behaved very similarly with virtually complete loss of soluble sugars, very high dry matter losses and a final pH value of about 8. All these bales had a similar black decomposed appearance.

TABLE 18

Comparison of Results of Mini-bale Experiment and First Batch Experiment

	Mini	Normal bales		Mini	Normal bales	
		High H_2O	Med. H_2O		High H_2O	Med. H_2O
Acid Level %	2.0	2.1	2.3	4.0	3.5	4.1
Initial Moisture Content %	51.1	45.5	34.3	50.2	41.6	39.6
Final Moisture Content %	23.9	33.7	28.3	43.4	33.2	29.4
Moisture Loss %	75.6	58.1	40.4	25.1	60.9	36.9
Dry Matter Loss %	25.7	10.4	12.4	3.4	8.9	2.6
W.S.C. Loss %	95.9	66.8	56.5	+10.2	66.3	40.1
Moulding %	100	60	35	0	35	20
Max. Temp.	34	51	29	18	44	29

In the first batch experiment however even the 1 and 2 per cent acid treatments did have some beneficial effect, which increased with the level of acid applied, and parts of the bales were found to be well preserved. It seems therefore that in the mini-bales when moulding started in one part of the bale it rapidly spread throughout as there were no areas which had a low enough moisture level, or a high enough acid treatment, to inhibit the mould. In the full size bales, however, it would seem that whatever the average acid treatment and moisture content there were always some areas of the bale which had a high enough treatment level or low enough moisture content, or combination of the two, to prevent the spread of mould to that part of the bale.

Although heating was clearly evident in the mini-bales which moulded, the temperatures reached were not as high as in the full sized bales. The mini-bales were kept in insulated boxes but these did not prove to be effective enough to compensate for the relatively large surface area of the mini-bales in comparison with full size bales. An indication of the heat output of the mini-bales is given by the very high moisture loss and the low final moisture content in the 2 per cent treated bales. That a large part of this 76 per cent loss was a result of heating is shown by a comparison with the 4 per cent treated bales in which no heating took place and only 25 per cent of the moisture was lost. By contrast the 2 per cent treated conventional sized bales, though reading a temperature 17^{o}C higher, lost only 58 per cent moisture, and finished with a moisture content 10 per cent higher than in the mini-bales. Again the smaller surface to volume ratio of the larger bales would restrict the rate of evaporation.

Dry matter losses also were very high in the mouldy mini-bales and it may be concluded that the actual temperatures reached are not of particular significance in themselves, the moulds continuing to grow until limited by other factors such as moisture content or nutrient supply. The lower temperatures in the mini-bales would be expected to affect the sequence and species of moulds and other organisms growing and becoming dominant on the hay. It is unlikely that, for example, thermophilic actinomycetes would be present to any great extent in the mini-bales as the optimum temperature for these organisms is about 50°C. However, such organisms do often grow prolificly in conventional sized bales which the hay is of a high moisture content (Gregory and Lacey, 1963a).

One other aspect of the results of the mini-bale experiment is worthy of discussion concerning the 1 and 2 per cent acid treatments which failed to preserve the hay. From the temperature graphs in Figure 29 it will be seen that the 1 and 2 per cent acid treatments progressively delayed the onset of heating and moulding so that the number of days taken before the maximum temperatures were reached increased from 7, in the untreated bales, to 14 in the 1 per cent bales and 25 in the 2 per cent bales. The higher acid levels prevented heating throughout the 14 weeks of the experiment. It is clear therefore, that as the level of propionic acid increases its inhibitory effect on growth of moulds becomes greater. Whether at the higher acid levels the moulds and mould spores are being killed or merely more effectively inhibited is not possible to say and is a question which has been debated without any convincing conclusions by a number of workers.

Again looking at the temperature graphs it will be seen that all

the acid treatments eliminated the initial temperature peak which is so clearly seen in the untreated bales. It would seem therefore that the propionic acid treatments lead to the rapid plasmolysis and death of the plant cells to which this initial temperature rise is normally attributed.

In conclusion it can be stated that as well as demonstrating clearly that mini-bales are a useful tool for studying the effect of propionic acid on hay under controlled conditions, the experiment has established that hay with a moisture content of 50 per cent can be satisfactorily preserved by an even application of propionic acid at 4 per cent by weight. Higher acid levels were shown to give no additional benefit, whereas treatments of 2 per cent and below were demonstrated to be insufficient to preserve hay at this moisture content.

SECOND MINI-BALE EXPERIMENT 1972

AIMS

The aim of this experiment was to establish the levels of propionic acid which would be required to preserve hay over a range of moisture contents. The first mini-bale experiment showed that mini-bales are a valid method for carrying out controlled experiments on hay. This second experiment was designed to be carried out in a similar way to the first, only at a number of moisture levels rather than at a single moisture level. In view of the comparative ease of producing mini-bales, it was decided to investigate a whole range of moisture levels from that contained in freshly harvested grass (Over 70 per cent) to that of nearly dry hay (30 per cent).

Although the first mini-bale experiment gave a clear difference between the well preserved hay treated with 4 per cent acid, and the hay treated with 2 per cent acid which went mouldy, there was no way of knowing if the 4 per cent treatment was only just adequate or whether a treatment level of just over the 2 per cent would have provided as good preservation as the 4 per cent level. To be of real value therefore, the second experiment would have to have much smaller steps of increasing acid level so that a more precise definition of the limits could be obtained. The increasing precision of course meant that the breadth of treatments applied would have to be limited otherwise the experiment would be of too large a size to be feasible.

Five acid treatments were chosen, at steps of $\frac{1}{2}$ per cent at each of the moisture levels 30, 40, 50 and 60 per cent. In each of these an untreated control was included. The acid treatments covered the range of treatments in which the 'critical' level for adequate

preservation was expected to be found as based on the results of the first mini-bale experiment and the two earlier batch experiments. It was impossible to predict what the effect of the acid would have on the fresh grass of very high moisture content and so wider range of treatments were adopted here with 1 per cent steps rather than $\frac{1}{2}$ per cent steps (Table 20).

Although this design is suitable for the comparison of different acid treatments at each moisture level, it leaves only a limited scope for the comparison of the effects of a single acid level at a number of moisture contents. Only with the control bales can a comparison be made over the complete moisture range while at the various acid levels a maximum of three moisture levels can be compared.

TABLE 20

Design of the Second Mini-bale Experiment

Moisture Level	Per cent Propionic acid by weight													
	0	0.5	1	1.5	2	2.5	3	3.5	4	4.5	5	5.5	6.5	7.5
Fresh grass	x							x		x		x	x	x
60%	x						x	x	x	x	x			
50%	x				x	x	x	x	x					
40%	x		x	x	x	x	x							
30%	x	x	x	x	x									

4 replicate bales at each treatment level arranged in randomised blocks

PLATE 5.

The Mark II mini-baler with a bag of loose hay and a mini-bale.

PLATE 6.

The mini-baler filled with loose hay, ready for the lid to be secured.

PLATE 7.

The mini-baler with the bale compressed and tied, ready to be released.

METHODS

Field and Storage Work

An area of about 1 acre of a pure perennial ryegrass stand was cut at the flowering stage. Freshly cut grass, sufficient to make the first set of mini-bales, was immediately taken to the store. The remainder of the grass was tedded and turned both by machine and by hand to promote rapid and even drying. Moisture contents were followed in the same way as previously described. As near as possible to each of the chosen moisture levels sufficient hay was taken to the store to make that set of mini-bales. In the store the hay at each moisture level was sampled, treated with propionic acid and made into mini-bales in exactly the same way as described for the first mini-bale experiment. Four replicate bales were made at each treatment level and the bales were as before stored in the insulated boxes of expanded polystyrene. The completed bales were again kept in an insulated potato store and arranged in four groups or blocks each of which contained one replicate from each treatment, arranged randomly.

The bales were stored over a three month period. Temperature readings were taken and the sampling and analyses were carried out in the same way as in the first mini-bale experiment.

The work of Shepperson (1956) has shown that the density of stored hay can have an effect on deterioration in that at higher the density the higher the temperature reached in the hay and the more rapid the moulding. This conclusion is confirmed in a separate small scale experiment with mini-bales, described in Appendix 13.

In preliminary trials for the experiment it was observed that

the hay of higher moisture contents packed more easily to a high density. Thus if all the mini-bales were to have been made at the same density of fresh matter then the dry hay would have been compressed to a greater pressure than that occurring in a normal baler, and the mini-baler would be used to the limits of its capabilities. In contrast, the wettest hay would have been so loosely packed as hardly to be classified as a bale.

Rather than make no attempt to make the bales of comparable densities, they were made in this experiment to have, as far as possible, the same dry matter density. In the results of the experiment the effect of density cannot be separated from the effect of moisture content and so it can only be said that every effort was made to keep the bales comparable in this respect.

The treatment of hay at above 50 per cent moisture content might be considered to be outside the realms of 'high moisture hay', being more like 'haylage' or wilted silage. Silage is, of course, made under anaerobic conditions, and so it was considered worthwhile to investigate whether, under aerobic conditions, propionic acid was capable of preserving such material. As no previous work had been done on this, there was no indication as to how much acid might be required. However, it was expected that the level of acid required for effective preservation would continue to increase with increasing moisture content as had occurred in the case in the 20 to 40 per cent moisture range in the first batch experiment.

RESULTS

All the moisture levels in the plan for the experiment were obtained with reasonable accuracy, as will be seen from Table 21. With the hay for the 60 and 50 per cent moisture levels the moisture content determination taken as the hay was brought in (quick drier) indicated a moisture content a few per cent above the intended level. The acid levels applied to these hays were stepped up accordingly. All the acid levels were therefore achieved very close to the original levels.

It may be noticed from the table that, at the 50, 60 and 76 per cent moisture levels the moisture contents of the untreated bales are uniformly 2 to 4 per cent higher than the hays in the propionic acid treated bales. Considering the fact that at each moisture level the treated and untreated bales were made from sub-samples of the same batch of hay, these differences seem strange. The possibility that the propionic acid applied, being volatile, has influenced of the oven dry matter determinations is considered in the discussion section.

The main results of the experiment will be presented in two sections, high moisture hays and low moisture hays.

High Moisture Hays

76, 60 and 50 per cent moisture contents

Tables 22, A, B & C

As would be expected at these moisture levels all the control, untreated bales showed severe rotting and decomposition. They were black in colour, had lost a considerable proportion of their original moisture due to the severe heating and were left with very little sol-

TABLE 21

Treatment Levels in the Second Mini-bale Experiment

Intended Moisture Level	Intended Acid Level	Achieved Moisture Level	Achieved Moisture Level Average	Achieved Acid Level
	%	%		%
Fresh Grass	nil	77.90	75.72%	nil
	3.5	75.30		3.64
	4.5	76.66		4.72
	5.5	74.86		5.64
	6.5	76.21		6.65
	7.5	73.40		7.63
60%	nil	61.42	58.92%	nil
	3.0	58.16		3.10
	3.5	58.99		3.60
	4.0	59.97		4.20
	4.5	56.63		4.60
	5.0	58.34		5.00
50%	nil	52.71	47.91%	nil
	2.0	47.16		2.20
	2.5	46.37		2.75
	3.0	46.75		3.21
	3.5	46.40		3.75
	4.0	48.06		4.20

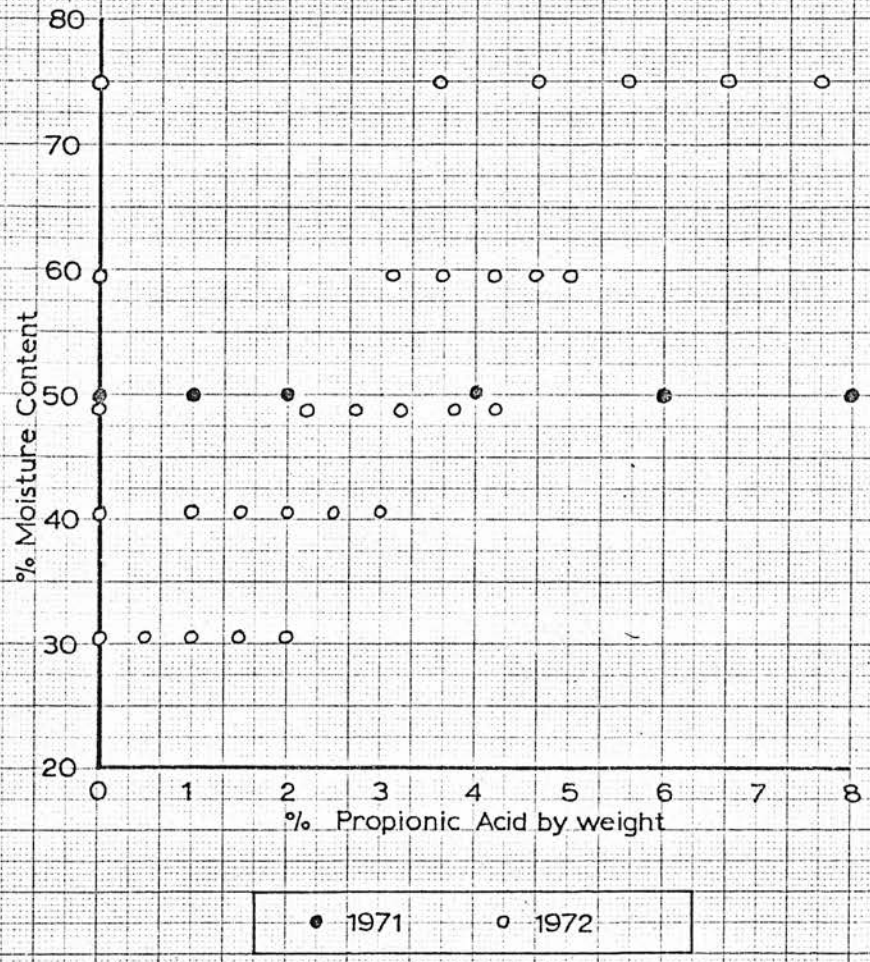
TABLE 21

ctd.

Intended Moisture Level	Intended Acid Level	Achieved Moisture Level	Achieved Moisture Level Average	Achieved Acid Level
	%	%		%
40%	nil	41.02	41.24 %	nil
	1.0	41.25		1.00
	1.5	40.50		1.50
	2.0	41.55		2.00
	2.5	42.10		2.50
	3.0	41.00		3.00
30%	nil	33.04	31.57 %	nil
	0.5	30.20		0.51
	1.0	30.40		1.00
	1.5	33.25		1.46
	2.0	30.98		2.04

Figure 29 B

MINI-BALE TREATMENTS 1971 & 1972



uble carbohydrate. Dry matter losses were highest at the 76 per cent moisture level, but heating was not so marked at this moisture level as it was in the untreated bales at the two lower moisture levels (Figure 30), in which the hay would have had a lower specific heat capacity due to the lower initial moisture contents. Most of the heating took place during the first 2 weeks of storage with a low initial peak in the first few days being followed by second more substantial period of heating.

All the propionic acid treatments applied to the bales at the three moisture levels reduced the temperatures to levels not significantly above the ambient (Figure 30) and no visible moulding was present in the bales. Dry matter, water soluble carbohydrate and moisture losses were significantly reduced in every case. At the highest moisture level, for example, dry matter losses fell from over 60 per cent to about 10 per cent, water soluble carbohydrate losses from 99 to between 17 and 40 per cent and moisture losses from 60 to 20 per cent. There were no significant differences between the losses at the different levels of acid application, however, at any of the moisture levels.

At the 50 and 60 per cent moisture levels the dry matter and soluble carbohydrate losses were, in most cases, not significantly different from zero, but in the 76 per cent moisture bales losses of both of these were much higher. Moisture losses in the treated bales at all three moisture levels were similar, amounting to about 25 of the original moisture content.

The pH values in the treated bales which showed no signs of moulding remained near to, or slightly below the initial pH. In the

TABLE 22A

MOISTURE LEVEL 76 per cent

Initial Water Soluble Carbohydrate 28.25%

							L.S.D.
Acid Treatment	nil	3.5	4.5	5.5	6.5	7.5	
Initial Moisture Content %	77.90	75.30	76.66	74.86	76.21	73.40	
Final Moisture Content %	78.56	72.39	72.26	72.15	73.53	71.97	
Initial pH	5.6	4.4	4.1	4.1	4.0	4.0	
Final pH	8.1	4.2	4.1	4.0	3.9	4.0	
Final Water Soluble Carbohydrate %	0.84	25.16	21.60	21.99	20.00	20.62	
Moisture Loss %	56.93	20.21	23.20	22.45	18.15	20.15	13.13
Dry Matter Loss %	61.22	7.90	3.30	11.79	6.16	15.00	3.61
W.S.C. Loss %	98.85	17.89	29.38	32.02	33.29	38.11	28.45
Degree Days above ambient	277.4	22.9	21.5	12.5	19.9	26.0	46.6
Visible moulding	severe	nil	nil	nil	nil	nil	

P = 0.01

TABLE 22B

MOISTURE LEVEL 60 per cent

Initial Water Soluble Carbohydrate 29.80%

							L.S.D.
Acid Treatment	nil	3.1	3.6	4.2	4.6	5.0	
Initial Moisture Content %	61.42	58.16	58.99	59.97	56.63	58.34	
Final Moisture Content %	54.65	55.07	56.18	54.97	53.51	56.05	
Initial pH	5.6	4.7	4.6	4.3	4.5	4.3	
Final pH	8.72	4.46	4.44	4.36	4.37	4.28	
Final Water soluble Carbohydrate %	0.69	25.74	29.68	29.96	25.45	30.07	
W.S.C. Loss %	98.85	20.44	9.54	4.41	24.78	10.72	29.2
Dry Matter Loss %	56.37	9.29	9.22	4.88	11.96	11.52	7.17
Moisture Loss %	65.47	19.92	18.95	22.37	22.07	19.17	47.41
Degree Days above ambient	374.3	29.9	-3.15	-0.5	-3.0	-3.5	38.5
Visible moulding	severe	nil	nil	nil	nil	nil	

P = 0.01

TABLE 22C

C. MOISTURE LEVEL 50 per cent — Initial Water soluble Carbohydrate 24.03%

							L.S.D.
Acid Treatment %	nil	2.2	2.8	3.2	3.8	4.2	
Initial Moisture Content %	52.71	47.16	46.37	46.75	46.40	48.06	
Final Moisture Content %	17.41	40.68	40.68	39.08	41.26	39.93	
Initial pH	6.50	5.20	4.95	4.79	4.80	4.59	
Final pH	8.00	4.65	4.62	4.59	4.55	4.60	
Final water soluble carbohydrate %	1.91	25.51	25.04	25.09	22.69	25.36	
W.S.C. Loss %	94.26	+2.41	+1.35	+9.05	10.33	+5.45	27.71
Dry Matter Loss %	29.39	3.74	2.91	0.94	5.25	0.10	5.71
Moisture Loss %	86.81	25.80	22.89	27.51	22.80	27.99	9.88
Degree Days above ambient	351.9	3.2	19.3	30.1	7.4	49.6	64.9
Visible moulding	severe	nil	nil	nil	nil	nil	

P = 0.01

TABLE 22D

D. MOISTURE LEVEL 40 per cent

Initial Water Soluble Carbohydrate 24.25%

							L.S.D.
Acid Treatment	nil	1.00	1.50	2.00	2.50	3.00	
Initial Moisture Content %	41.02	41.25	40.50	41.55	42.10	41.00	
Final Moisture Content	19.38	24.18	31.93	32.03	31.79	32.14	
Initial pH	6.00	5.60	5.30	5.19	4.99	4.89	
Final pH	8.21	7.48	5.12	4.94	4.89	4.74	
Final water soluble Carbohydrate %	4.18	4.13	16.55	17.94	19.92	17.04	
W.S.C. Loss %	88.05	86.10	31.96	25.18	16.13	30.27	29.66
Dry Matter Loss %	30.17	17.60	0.44	+1.03	+2.15	0.77	5.38
Moisture Loss %	75.88	62.44	31.01	33.00	34.52	32.16	3.20
Degree Days above ambient	220.2	91.5	34.6	35.9	28.2	34.1	48.2
Moulding visible	severe	moderate	nil	nil	nil	nil	

P = 0.01

TABLE 22E

E. MOISTURE LEVEL 30 per cent

Initial Water Soluble Carbohydrate 24.90%

						L.S.D.
Acid Treatment	nil	0.51	1.00	1.46	2.04	
Initial Moisture Content %	33.04	30.20	30.40	33.25	30.98	
Final Moisture Content %	18.80	18.24	24.64	24.53	24.24	
Initial pH	6.10	5.90	5.75	5.51	5.31	
Final pH	7.32	6.66	5.71	5.61	5.23	
Final water soluble carbohydrate %	3.32	6.54	16.91	16.19	19.44	
W.S.C. Loss %	92.09	78.07	36.36	35.16	24.03	34.18
Dry matter loss %	21.40	18.66	6.41	0.20	2.65	9.16
Moisture loss %	63.15	57.95	29.96	34.84	26.02	4.86
Degree days above ambient	116.2	78.9	29.6	18.7	13.9	37.6
Visible Moulding	severe	moderate	slight	nil	nil	

P = 0.01

untreated bales the pH value rose from about 6 to over pH 8 in each case.

Low Moisture Hays

40 and 30 per cent moisture contents

TABLE 22 D+E.

Once again the untreated bales showed severe moulding. At the 30 per cent moisture level the mouldy bales had a yellow appearance indicating that the predominant moulds were of the Aspergillus glaucus group which produce large numbers of yellow perithecia and amass as a yellow dust on and beneath the bales. The untreated bales at 40 per cent moisture turned a blackish colour. In all the untreated bales the loss of water soluble carbohydrate was again almost complete, and dry matter losses were high, being 30 per cent in the 40 per cent moisture hay and 21 per cent in the 30 per cent moisture hay. Heating amounted to 220 day degrees and 116 day degrees in the 40 and 30 per cent moisture hays respectively.

The temperature graphs in Figure 31 show that very little heating took place during the first two weeks of storage, but when there was a slight rise in the ambient temperature at the end of the second week the untreated bales began to heat more rapidly, especially at the 40 per cent moisture level and continued to maintain temperatures above the ambient until about 6 weeks after the start of storage.

The lowest propionic acid treatment levels at both the 40 and the 30 per cent moisture contents, 1.0 and 0.5 per cent respectively, were unable to prevent moulding in the bales. Although in both cases the acid delayed the onset of heating (Figure 31) and reduced its overall extent, considerable moulding was present in all the bales. Water

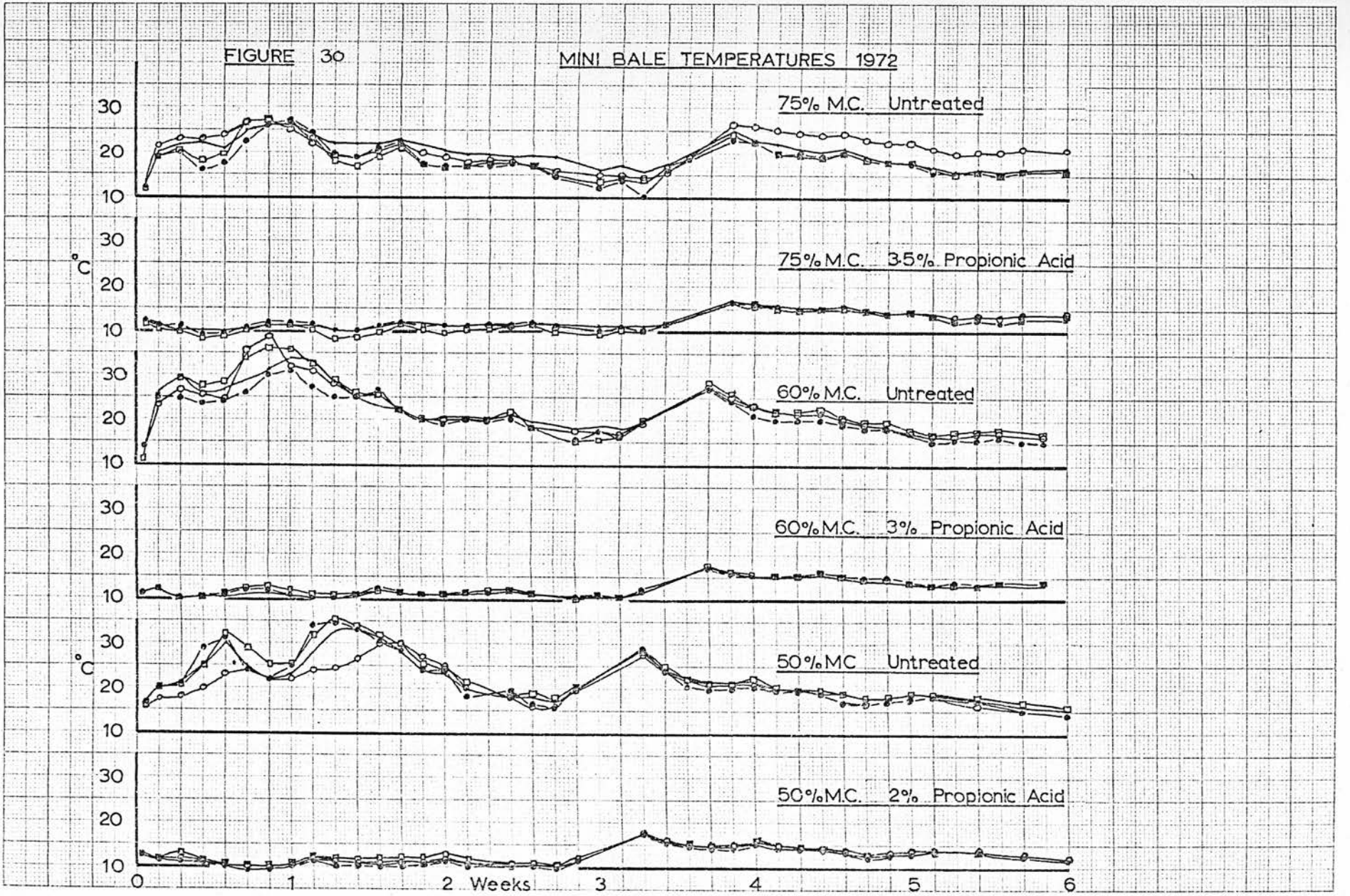
soluble carbohydrate losses were not significantly different than in the untreated bales, although the dry matter loss in the 40 per cent moisture hay was. Moisture losses follow a similar pattern to the heating, and were significantly reduced even by these lowest acid levels.

The propionic acid levels of 1.5 per cent and above on the 40 per cent moisture hay, and 1.0 per cent and above on the 30 per cent moisture hay completely, or almost completely, prevented visible moulding on the bales. Heating in each case was not significantly above zero and was less than that in the bales of the lowest acid levels. Dry matter losses were insignificant at both moisture levels, although water soluble carbohydrate losses tended to be rather high, in the order of 30 per cent. In all these well preserved bales the losses were significantly lower than in the untreated bales and the bales which received the lowest acid treatment levels.

As in the high moisture hays the differences between losses at the various acid levels, not including the lowest ones, were small and insignificant. In one of the 30 per cent moisture bales which received the 1.0 per cent acid treatment, however, some visible moulding was present, but was not of sufficient extent to give any noticeable temperature rise, or any increase in the losses.

The pH values also followed the same pattern as those of the high moisture hays, with the pH of the well preserved bales remaining the same or falling slightly while in the bales which showed moulding the pH rose during storage.

FIGURE 30


MINI BALE TEMPERATURES 1972
75% M.C. Untreated
75% M.C. 3.5% Propionic Acid
60% M.C. Untreated
60% M.C. 3% Propionic Acid
50% MC Untreated
50% M.C. 2% Propionic Acid
°C
30
20
10
0
1
2 Weeks
3
4
5
6

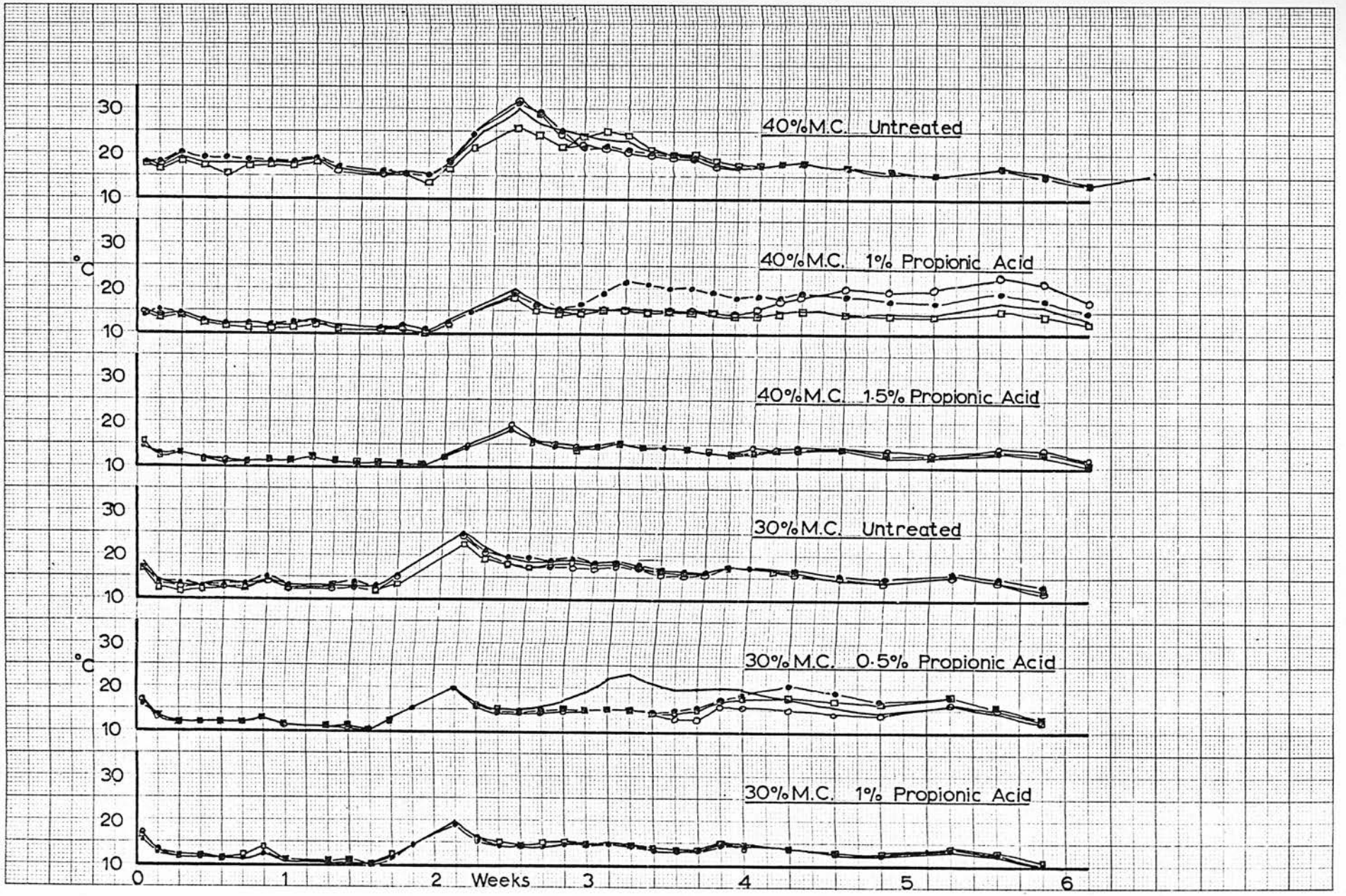

40% M.C. Untreated
40% M.C. 1% Propionic Acid
40% M.C. 1·5% Propionic Acid
30% M.C. Untreated
30% M.C. 0·5% Propionic Acid
30% M.C. 1% Propionic Acid
°C
30
20
10
0
1
2
Weeks
3
4
5
6

PLATE 8.

Moulding in an untreated high moisture batch in the first batch experiment

PLATE 9.

Mini-bales showing different levels of moulding in the second mini-bale experiment

DISCUSSION

In this experiment a very wide range of moisture contents were encompassed. While the lower moisture levels were within the normal scope of hay, grass at the higher moisture levels of 60 to 80 per cent would normally be stored anaerobically to make fresh or wilted silage. Indeed, propionic acid at low application rates has been used as an experimental silage additive under these conditions (Daniel *et al*, 1970). There is no record, however, of experimental work with propionic acid as a preservative for such high moisture material stored under aerobic conditions and so it was difficult to predict even approximately what effect the acid would have under the conditions of this experiment.

The treatment levels chosen for the experiment were based on the conclusion from the 1971 mini-bale experiment that at 50 per cent moisture a treatment of between 2 and 4 per cent acid is needed for adequate preservation. It was also assumed that as the moisture content fell, so the acid requirement would fall until, at a moisture level of about 20 per cent the hay would not require acid for preservation. Experimental work has shown that some mould growth will be present on hay down to 15 per cent (Waite 1949) but it is only above about 20 per cent moisture in bales that it will take place to any appreciable extent (Watson and Nash, 1960 C). Above moisture contents of 50 per cent there were no previous results to go on.

Since the aim of the experiment was to establish the level of acid which would be just adequate for preservation at each moisture level this aspect will be considered first. Over the whole experiment the range of acid treatments chosen tended to be on the high side and this

is particularly noticeable at the three highest moisture levels where even at the lowest levels of acid no moulding occurred. The reason for this becomes apparent if the lowest acid level, 2.2 per cent, on the 48 per cent moisture hay is considered. This 2.2 per cent acid treatment effectively preserved the hay whereas, in the first mini-bale experiment the 2.0 per cent acid treatment on hay of 51 per cent moisture did not prevent moulding (Figure 29). Thus it seems clear that 2 per cent acid may be on the border line between an adequate and an inadequate treatment for hay of about 50 per cent moisture. This tentative conclusion is based on the assumption that the first and second mini-bale experiments are comparable, since the losses in the untreated bales for each of the mini-bale experiments are very similar (Tables 22C & 17).

At the lower moisture levels, however, there is a clearer indication of where the limits of an adequate treatment might lie, for the lowest level of acid in each case showed mouldy deterioration, whereas those 0.5 per cent acid higher did not deteriorate, except slightly in one out of the four bales at 30 per cent moisture. Thus the level for adequate preservation would appear to lie just below 1 per cent propionic acid on hay of 30 per cent moisture, between 1.0 and 1.5 per cent on hay of 40 per cent moisture, and at 2 per cent acid for hay of about 50 per cent moisture.

In Figure 33 a comparison can be made of the treatment levels which prevented moulding and those which did not, taking both the mini-bale experiments together. On the graph a line has been drawn to indicate the possible lower margin of propionic acid application levels which would effectively preserve hay at the various moisture levels. The origin of the line has been taken to be the 15 per cent moisture

level at which Waite (1949) claims that all mould growth ceases on hay. At each moisture level the line passes through, or nearly through, the lowest adequate acid treatment level. Above the 50 per cent moisture level however, none of the acid treatment levels were inadequate and so it is not possible to say whether the lowest acid levels were in fact anywhere near the limit of effective preservation. Assuming that the graph continues as a straight line, however, the results would suggest that the lowest acid levels applied to the high moisture hays were in fact near the lower limit for effective preservation.

The line of minimum adequate preservation may be represented by the equation:-

$A = (M - 15) \times 0.06$ where A = Propionic acid treatment, per cent by weight

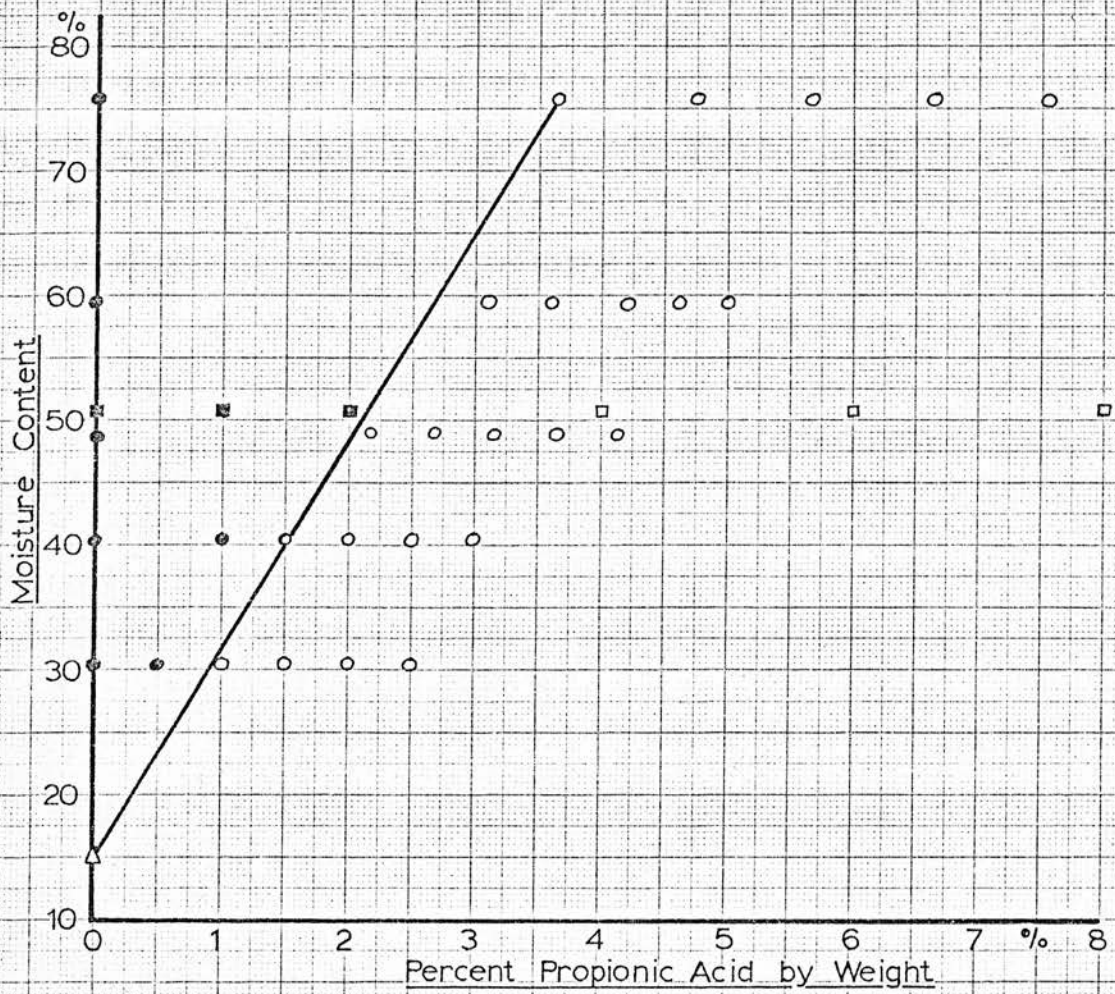
M = Moisture content, per cent of fresh weight

Whether it is legitimate to extrapolate the graph as a straight line above the 50 per cent moisture level is an open question, however, as the fresh grass is such an entirely different material from partly dried hay.

In the mini-bales made from the fresh grass, water soluble carbohydrate and dry matter losses were significantly above zero, although the bales seemed well preserved and the pH did not rise. A similar picture emerges at the 60 per cent moisture level, although water soluble carbohydrate losses were lower, while at the 50 per cent moisture level the losses of water soluble carbohydrate and dry matter were insignificant.

At the two highest moisture levels a bacterial fermentation could

FIGURE 33 Minimum Adequate Level of Propionic Acid Treatment for the Preservation of Moist Hay



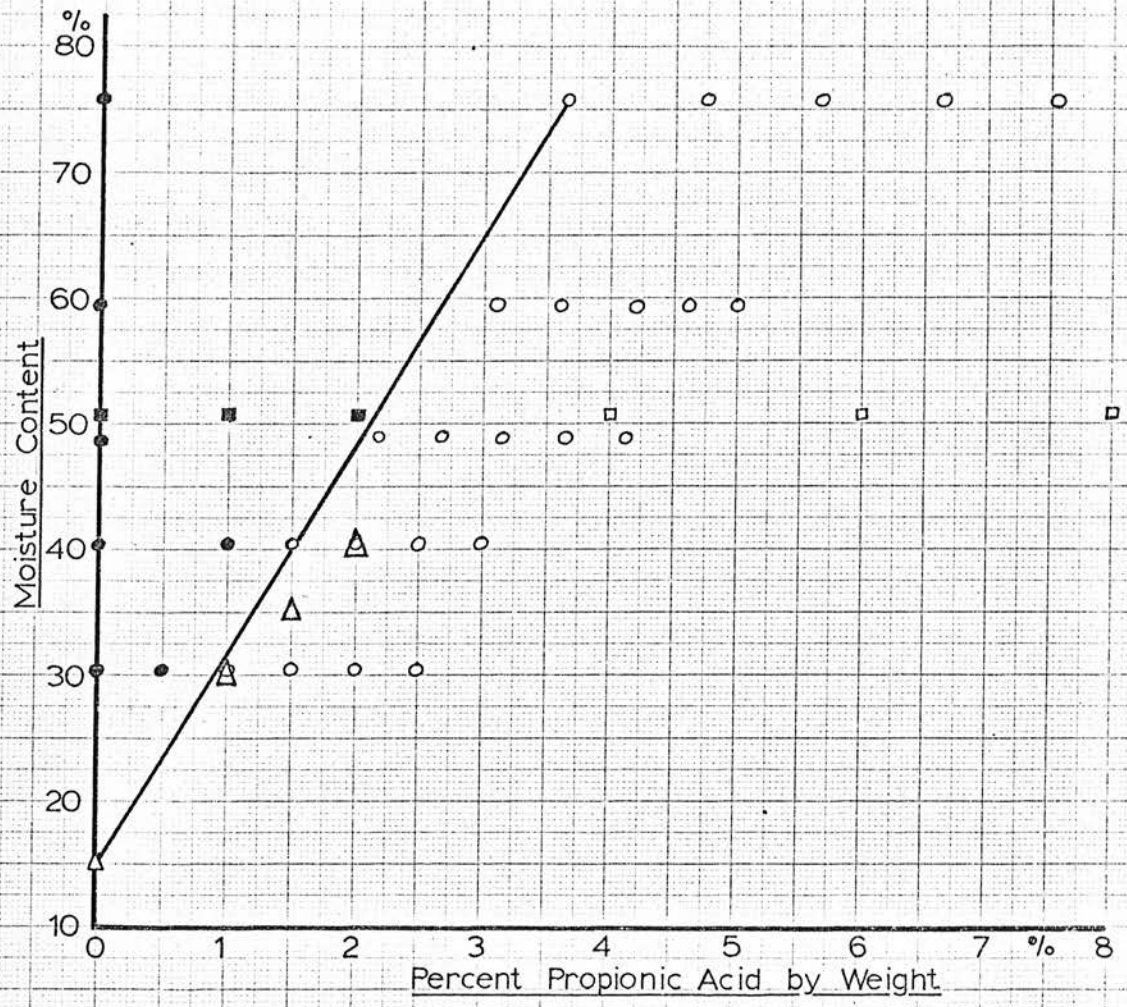
MINIMUM ADEQUATE TREATMENT LEVEL

Minimum Moisture Content at which Moulding will take place. (Waite, 1949)

		mouldy	non-mouldy
Mini-bales	1971	■	□
Mini-bales	1972	●	○

FIGURE 34 Minimum Adequate Level of Propionic Acid Treatment for the Preservation of Moist Hay

(Including the results of Shukking, 1972)



MINIMUM ADEQUATE TREATMENT LEVEL

Minimum Moisture Content at which Moulding will take place. (Waite, 1949)

		mouldy	non-mouldy
Mini-bales	1971	■	□
Mini-bales	1972	●	○
Shukking	1972		Δ

have aided the propionic acid in the preservation of the material. If this had taken place it would help to explain why there was a significant loss of water soluble carbohydrate in most of the treated bales ranging from 4.41 to 38.11 per cent (Tables 22A & B). The high moisture content is less favourable to mould growth, whereas it is more suitable for the growth of bacteria and yeasts which grow best in an almost saturated environment. It is interesting to note that when the acid treatments were applied, the pH of the fresh grass in this experiment was reduced to 4.4 with the 3.5 per cent acid treatment and to as low as 4.1 by the higher acid treatments. In silage such concentrations of hydrogen ions would readily inhibit the activity of most bacteria other than the lactic acid types, which under the continuing anaerobic conditions of good silage making may tend to lower the pH even further, by producing lactic acid at the expense of water soluble carbohydrate. Lactic acid bacteria are known to multiply also under aerobic conditions, as they are facultative anaerobes, but their by products under these conditions have not been fully investigated.

If lactic acid bacteria or others had been respiring aerobically during storage this would have brought about oxidative changes leading to a rise in the pH value. However, a pH rise was not observed and so bacterial oxidation is unlikely to have taken place.

Evidence of the occurrence of an anaerobic fermentation would have been the presence of fatty acids other than the propionic acid applied to the hay. But traces of acid other than propionic acid were not found when samples were analysed by separation in a silica gel chromatographic column. The final pH of the high moisture treated hays was slightly lower after the storage period than before it, but this trend was also present in the low moisture hays which would

indicate that it was not due to a fermentation. It may have been due to chemical changes in the natural buffering agents present in grass, during the storage period.

The relatively high losses of water soluble carbohydrate in many of the well preserved bales are not easy to explain if bacterial fermentation and mould growth are entirely discounted. Furthermore, it was not only at the highest moisture level, 76 per cent, but also in the well preserved mini-bales at the lowest moisture level, 30 per cent, that losses of as high as 36 per cent were recorded.

One possible source of soluble carbohydrate loss, but only at the 76 per cent moisture level, was a small quantity of effluent, about 100 cm^3, at the maximum, which was expressed from the grass each time a mini-bale was made. This loss only took place from the bales which were being made from acid treated grass and is likely to have been due to the rapid death of the plant cells on treatment with propionic acid. A similar increased flow of effluent has been noted in experimental work where silage has been treated with formic acid (Pedersen *et al*, 1973). However, the quantity of effluent lost in this experiment only amounted to about 100 cm^3 which would be approximately 5 per cent of all the water initially present in the bale. Assuming that the water soluble carbohydrate would not be at a greatly increased concentration in one part of the bale it would be unlikely that more than 5 per cent of the initial water soluble carbohydrate could be lost with the effluent and it would not seem to give a fully satisfactory way of explaining the overall loss of 30 per cent of the sugar.

While these losses may have actually occurred, it is possible that they did not in fact take place but appear in the results as a

consequence of a weakness in the method used to calculate the losses. The most likely source of error would be the sampling of the hay to provide the initial water soluble carbohydrate determination, the losses in all the bales at one moisture level being based on a single result, the mean of a triplicate determination. If the original sample was not truely representative then the final result would be an underestimate or overestimate of the actual losses. While every precaution was taken to make sure that the samples used were representative by thorough mixing of the hay before and during sampling, the possibility of error from this source remains.

In the low moisture hay at 30 per cent moisture, for example, the initial water soluble carbohydrate level was 24.9 per cent which was slightly more than that in the 50 per cent moisture hay. However, the 30 per cent moisture hay had lain in the field 8 days longer and it would normally be expected that this would result in a loss of water soluble carbohydrate by oxidation or leaching. It is possible therefore that the initial water soluble carbohydrate value for the 30 per cent moisture hay is an overestimate which would explain the exceptionally high losses which appeared to have occurred during storage.

Whether or not these high water soluble carbohydrate losses did take place, it does not question the fact that these bales were otherwise well preserved with no appearance of visible moulding.

While this experimental work was in progress the results of similar work being carried out by Shukking in Holland were released (Shukking, 1972). In laboratory scale trials using insulated boxes about 25 cm^3 he found that at 31, 40 and 50 per cent moisture 0.8, 1.5 to 2 and 2.5 to 3 per cent propionic acid respectively were required

to preserve that hay over a 30 day period. No details are given about the materials and methods used in these experiments and the storage period would seem short for full confidence to be placed in the results. However, when the results are plotted against those of the present mini-bale experiments it will be seen that they are in reasonable agreement (Figure 34).

It may be concluded therefore that this experiment using mini-bales has been successful in establishing with reasonable confidence the minimum levels of propionic acid required to preserve hay over a range of moisture contents between 30 and 50 per cent. The experiment was carried out under conditions in which the propionic acid was applied evenly to the hay which had a uniform moisture content and so the results could not be used as recommended levels for the preservation of hay made and treated with propionic acid in the field under conditions in which neither the propionic acid application nor the moisture content of the hay is uniform.

At moisture contents above 50 per cent the experiment has shown that propionic acid, if supplied at sufficiently high levels, can adequately preserve partially dried or even fresh grass without moulding or bacterial fermentation. Evidence of dry matter and water soluble carbohydrate losses from such material was found, however, although the source of the losses could not be definitely established.

SECTION THREE

FIRST DISTRIBUTION EXPERIMENT

AIMS

While it is clear that propionic acid can effectively preserve moist hay, a number of problems are encountered when an attempt is made to use this method for preserving hay 'in the field'. The photographs on pages 45 & 234 show typical swaths of hay (10) ready for baling and (1) being baled and at the same time treated with propionic acid. The hay in (10) can be seen to lie unevenly in the swath because of the tendency of the almost dry hay to become entangled into lumps during tedding and turning, leaving parts of the swath almost bare.

Although the acid will emerge from the applicator at an even rate and in the form of a fine mist, which will penetrate some of the hay as it is picked up by the baler, the unevenness of the swath itself is bound to lead to variation in the amount of acid reaching different portions of the hay as it is baled.

By the end of the First Batch experiment it was clear that mouldy patches would develop in bales, even when very high overall rates of acid were applied. For example in the hay with a moisture content of 36 per cent, mould patches developed even when an overall acid level of 4.1 per cent was applied, while in the later mini-bale experiments it was shown that under ideal conditions an acid level of under 1.5 per cent would be adequate to preserve hay at this moisture level. It seemed highly probable, therefore, that uneven acid distribution due to the condition of the swath at the time of baling was a factor contributing towards the eventual appearance of moulding in the acid treated bales.

Another factor contributing to the mould patches is likely to have been a variable moisture content in the swath. Gregory et al (1963) noted that hay baled at 30 to 35 per cent moisture was unevenly dried, and other workers have noted that wet patches can occur if swaths are not handled properly (Mitchell and Shepperson, 1955). Frequent tedding and turning of the swath has always been advocated for the reason that drying will take place more quickly on the top of the swath which is open to the wind and sun. Where parts of the swath are thicker than others it would be expected that the thicker parts would dry more slowly and that the centre of large lumps could remain quite damp in spite of repeated teddings and turnings.

Where the hay is going to be finished in small stacks in the field or by barn drying, slight irregularities in the moisture content will not be of importance for they will be evened out as the hay continues to lose moisture. Thus the methods of haymaking in current use do not lay a particular stress on producing an even swath and evenly dried hay. However, when an acid treatment is used to preserve the hay irregularities become of vital importance for the reasons already outlined.

The need for even swaths of hay and even distribution of the acid through the hay can be appreciated without further consideration, but the questions requiring answers are:-

a. How great are the variations in moisture content and thickness along a 'typical' length of swath?

b. How great are the variations in propionic acid treatment within a bale?

c. Which of these factors is the more important in causing the patches of mouldy hay found in treated bales?

d. To what extent is applicator design responsible for variations in the propionic acid treatment reaching the hay?

The aim of this experiment and of the Second Distribution Experiment is to try and answer these questions by making a close examination of swaths and of bales made from these swaths. The first experiment is concerned with a single length of 'normal' swath and with the bales made from it. This forms a basis for the second experiment in which four types of swath are artificially prepared and the bales from these studied in detail.

DESIGN

A detailed examination was made of a randomly selected 155 metre length of swath. Measurements were made of the weight of hay, maximum height and maximum width of the swath for each metre length. Each metre was then labelled and numbered consecutively so that after baling the exact parts of the swath from which each bale had been made could be identified. The swath was baled and treated with propionic acid using the Low Pressure applicator supplied by BP Chemicals (International) Ltd. (Appendix 7).

Immediately after baling a number of bales from the swaths were dissected into 40 small parts, taking particular note of the labels found which identified the parts of the swath from which the bale had

come. Each of the 40 parts was then analysed for moisture content and propionic acid level. The variations within the bales could then be compared with those in the part of the swath from which the bale had been made and any link between them established.

METHODS

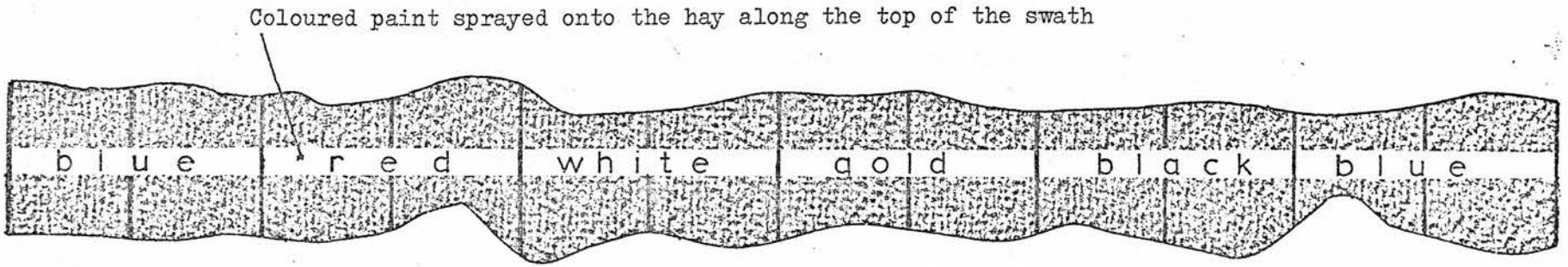
Field Work

A field of perennial ryegrass was cut at the flowering stage and field dried in accordance with the normal practice for rapid haymaking. Moisture contents were taken as outlined in the first batch experiment. When the moisture content was in the 30 to 40 per cent range the swaths were windrowed (two swaths run into one) in readiness for baling.

A length of the windrowed swath of about 155 metres was allocated for the experiment and, divided into 1 metre lengths using a pair of shears to cut the hay and provide a clean division, but without leaving a gap. The maximum width and height of each metre length were measured. The weights were taken one by one by rolling each portion of hay onto a polythene sheet which was then hooked onto a spring balance and weighed to the nearest 50 grams. After weighing, each portion of hay was carefully replaced and left in as nearly as original physical condition as possible.

Numbered plastic labels were loosely placed in the hay in the centre of each metre length of hay. Aerosol sprays of paint were used to provide permanent identification of the different lengths of the swath. Five colours were sprayed in sequence in a 4 inch broad band

FIGURE 35 CONDITION OF SWATH AT TIME OF BALING



along the middle line of the swath, each colour being used over a 2 metre length of swath. The sequence of colours was:- Blue; red; white; gold; black. Figure 35 shows the condition of the swath immediately prior to baling.

As soon as the measurements and marking had been completed baling took place; four untreated bales followed by eight bales treated with propionic acid. Two rates of propionic acid application rate were used, 2 per cent and 1.5 per cent approximately, four bales being made at each level. The acid was applied using the Low Pressure Applicator, Mark I (Appendix 7).

From these 12 bales four were randomly selected for immediate analysis, namely 2 untreated, 1 high acid and 1 low acid bale.

Bale Analysis

The method of dividing the bale is illustrated in Figure 36. Each bale was first divided into 8 sections of as nearly equal thickness as possible, labelled A to H. Four of these sections, chosen alternately, were further divided, using a power driven band saw, into 9 equal parts. In the first of the untreated bales only two of the sections were divided in this way.

The hay from the remaining sections and all the individual parts was chopped into short lengths (about 1 cm) using a small 'Bentall' straw chopper. As the hay was chopped the number tags were retrieved and their location in the bale noted. Any colour marks in the hay were also noted. Duplicate oven dry matter determinations were carried out on all the sections and parts of the bale.

Estimations of propionic acid levels formed a vital part of this

investigation, but no method was available by which the level of propionic acid on the hay could be measured directly, without time consuming procedures which would have severely restricted the number of samples which could have been dealt with in the time available.

One of the most easily determined factors affected by the level of propionic acid applied is the pH of the hay, and so after a preliminary investigation into the effect of propionic acid on the pH it was decided to use this parameter to estimate the level of acid on each sample of hay.

In order to provide standards against which the hay from the bales could be compared a quantity of hay was taken from the field at the time of baling and treated in the laboratory with an accurate range of acid treatments by means of a hand sprayer. The range of acid treatments used was from 0 to 7 per cent propionic acid, by weight. The pH was then determined in the same way as with all the subsequent samples, by macerating 10 grams of the chopped hay sample with 200 ml. of deionised water for 2 minutes and measuring the pH with an electronic meter. The results when plotted as a graph of pH against acid treatment are shown in Figure 37, from which the level of propionic acid on a sample of hay can be directly estimated from the pH value.

This method of estimation suffers from the obvious disadvantage that, as the acid level increases the relative change in pH becomes smaller and smaller, and so the accuracy of the estimate becomes less and less. This feature is not only due to the low dissociation constant of propionic acid, but also to the buffering agents in grass which tend to oppose any pH changes. This method may therefore be considered to be poor for estimating high levels of propionic acid;

FIGURE 36

FIRST DISTRIBUTION EXPERIMENT

BALE ANALYSIS

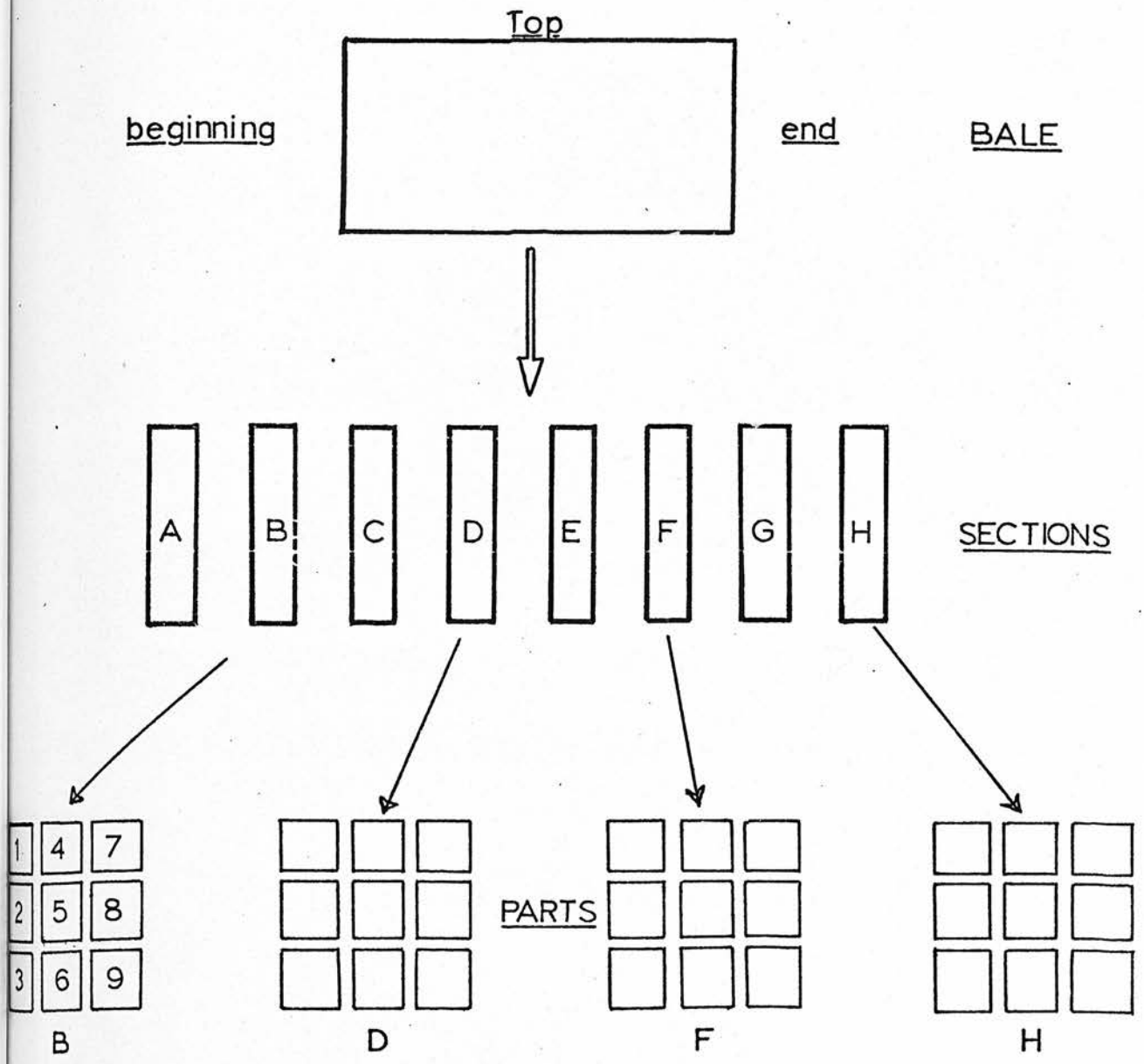


Figure 37

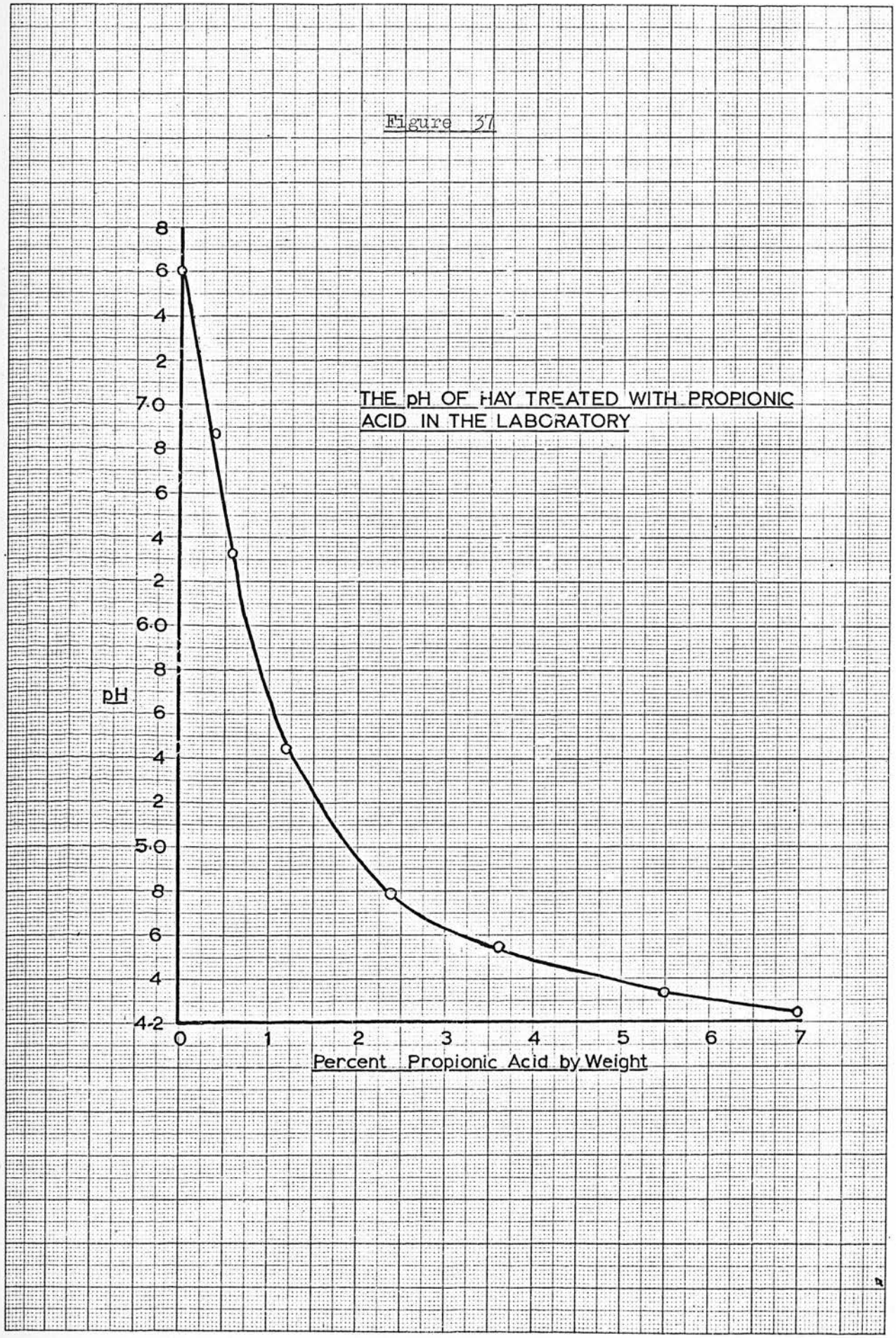
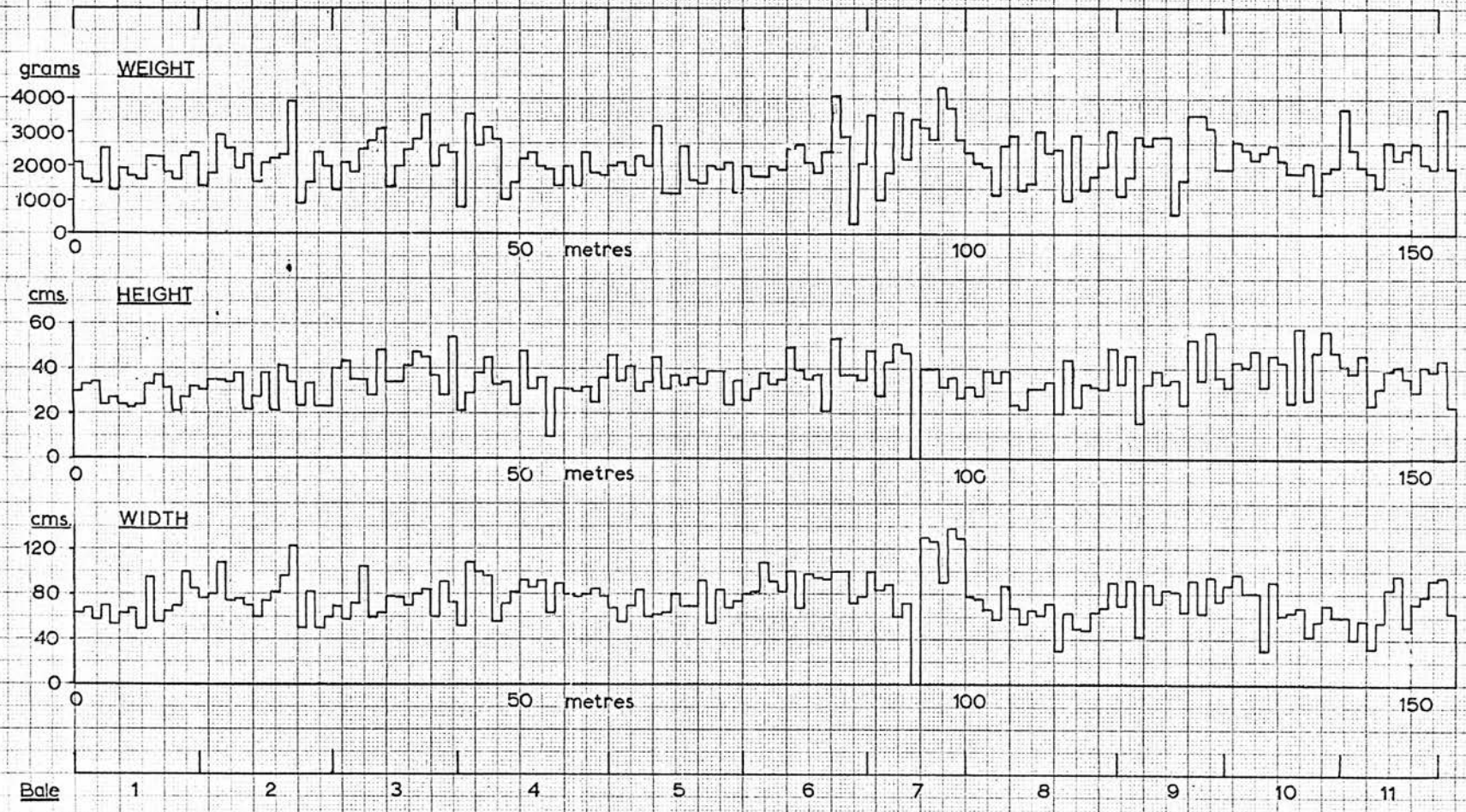

THE pH OF HAY TREATED WITH PROPIONIC ACID IN THE LABORATORY
pH
8
6
4
2
7.0
8
6
4
2
6.0
8
6
4
2
5.0
8
6
4
4.2
0
1
2
3
4
5
6
7
Percent Propionic Acid by Weight

FIGURE 38 SWATH WEIGHT, HEIGHT AND WIDTH PER METRE LENGTH



above about 5 per cent by weight. However, the method was considered to be sufficiently accurate for the purpose of this preliminary investigation.

RESULTS

The measurements taken on the swath before baling are represented graphically in Figure 38. The figure also shows the lengths of swath comprising the first 11 bales in the experiment. In order that a valid comparison can be made between the different measurements the coefficient of variation has been used, which is the standard deviation of the values expressed as a percentage of the mean.

$$\text{Standard Deviation} = \frac{\text{Sum of squares of deviations from the mean}}{\text{number of values}}$$

$$\text{Coefficient of variation} = \frac{\text{Standard deviation}}{\text{mean}} \times 100$$

Table 23 — Measurement on the Whole Swath

Whole Swath (115 metres)	Mean	Standard deviation	Coefficient of Variation
Swath weights	2205g	718.9	32.60
Swath heights	34.7cm	9.1	26.22
Swath widths	76.2cm	19.2	25.20

From the above table it can be seen that of the three series of measurements, the swath weights were the most variable, with an almost 33 per cent variation. From Figure 38 it will be seen that this

represents a range of values from under 200 g to over 4000 g in the 155 measurements. Although the variations in height and width were not so marked they still amounted to more than a 25 per cent variation.

The pattern of the values in Figure 38 do not show any particular trends except that low values tend to be preceeded or followed by above average values, this trend being evident in all three parameters. The correlations between the weights, heights and widths were small, being in the order of 0.2 to 0.5 indicating that the height and width of the swath was only dependent to a small extent on the weight of hay in each metre length.

With the aid of the numbered labels and coloured markings the sections of the swath from which each bale was made were able to be located. In Figures 39, 40 and 41 the swath weights for the length of swath from which each bale was made are presented together with the moisture contents and propionic acid levels found in the various sections and parts of the bales. Further information is given in Table 24 which shows the means, standard deviations and coefficients of variation for the four bales.

The table of mean values shows that the four bales had mean moisture contents of between 34 and 40 per cent and that the two acid treated bales received 1.52 and 2.14 per cent acid respectively. As would be expected the moisture content value derived from the sections of the bales agreed quite closely with those obtained by meaning the values for the smaller parts of the bales (see Table 24). Although the means were similar, the moisture contents in the smaller parts were significantly more variable than those in the larger sections and over the four bales ranged from under 30 per cent to over 50 per cent.

Table 24

Results of Bale Analyses

Bale number	Untreated 2	Untreated 3	Acid treated 5	Acid treated 10
Means				
Swath weights, grammes	2106	2269	1917	2185
Moisture contents (parts) %	35.75	39.14	36.53	35.54
Moisture contents (sections) %	34.30	38.16	36.84	34.68
Acid Levels (parts) %	-	-	1.84	2.12
Acid Levels (sections) %	-	-	1.52	2.14
Standard Deviations				
Swath weights	665	553	627	434
Moisture contents (parts)	4.89	3.72	2.72	4.49
Moisture contents (sections)	3.20	3.32	1.62	2.59
Acid Levels (parts)	-	-	0.76	0.70
Acid Levels (sections)	-	-	0.27	0.32
Coefficient of Variation				
Swath weights	31.58	28.87	27.65	19.84
Moisture contents (parts)	13.68	9.51	7.45	12.63
Moisture contents (sections)	9.33	8.70	4.39	7.42
Acid Levels (parts)	-	-	41.55	33.30
Acid Levels (sections)	-	-	18.03	15.08

FIGURE 39 1971 BALE ANALYSIS BALE 3

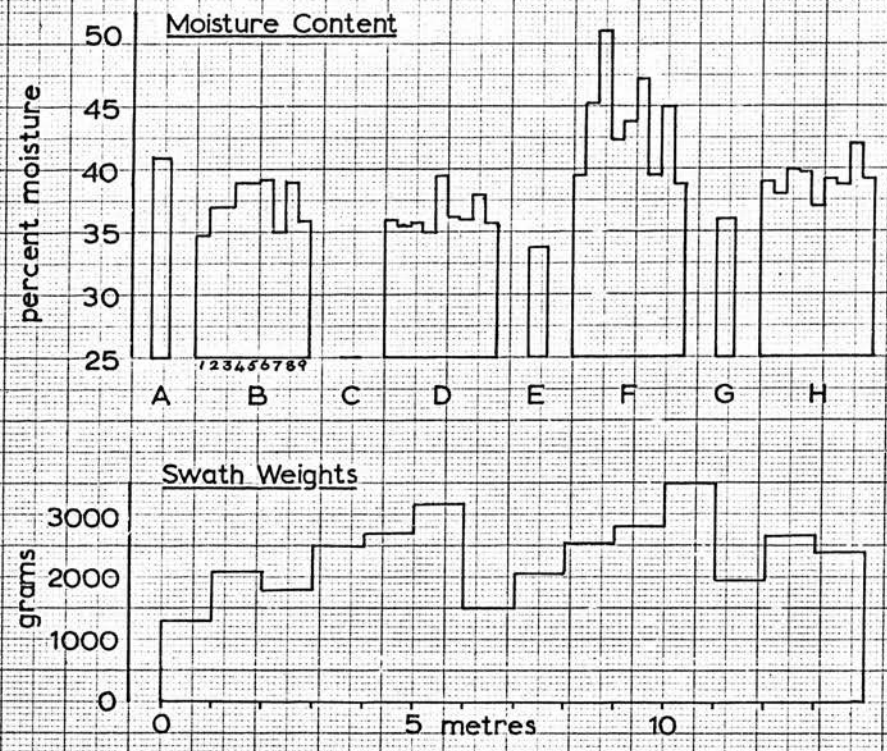


FIGURE 1971 BALE ANALYSIS BALE 2

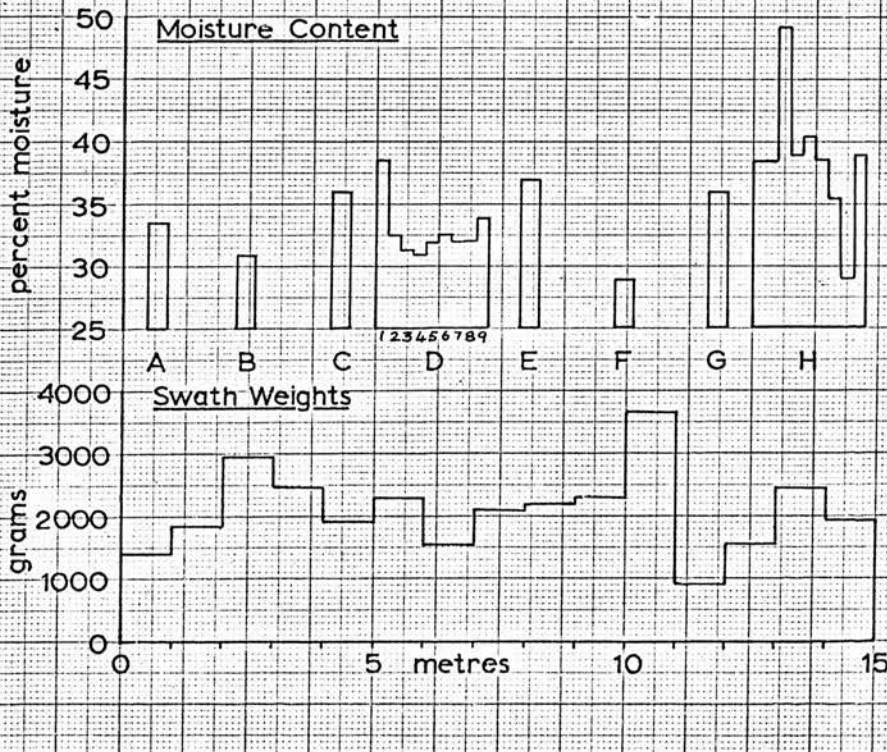


FIGURE 40 1971 BALE ANALYSIS BALE 5

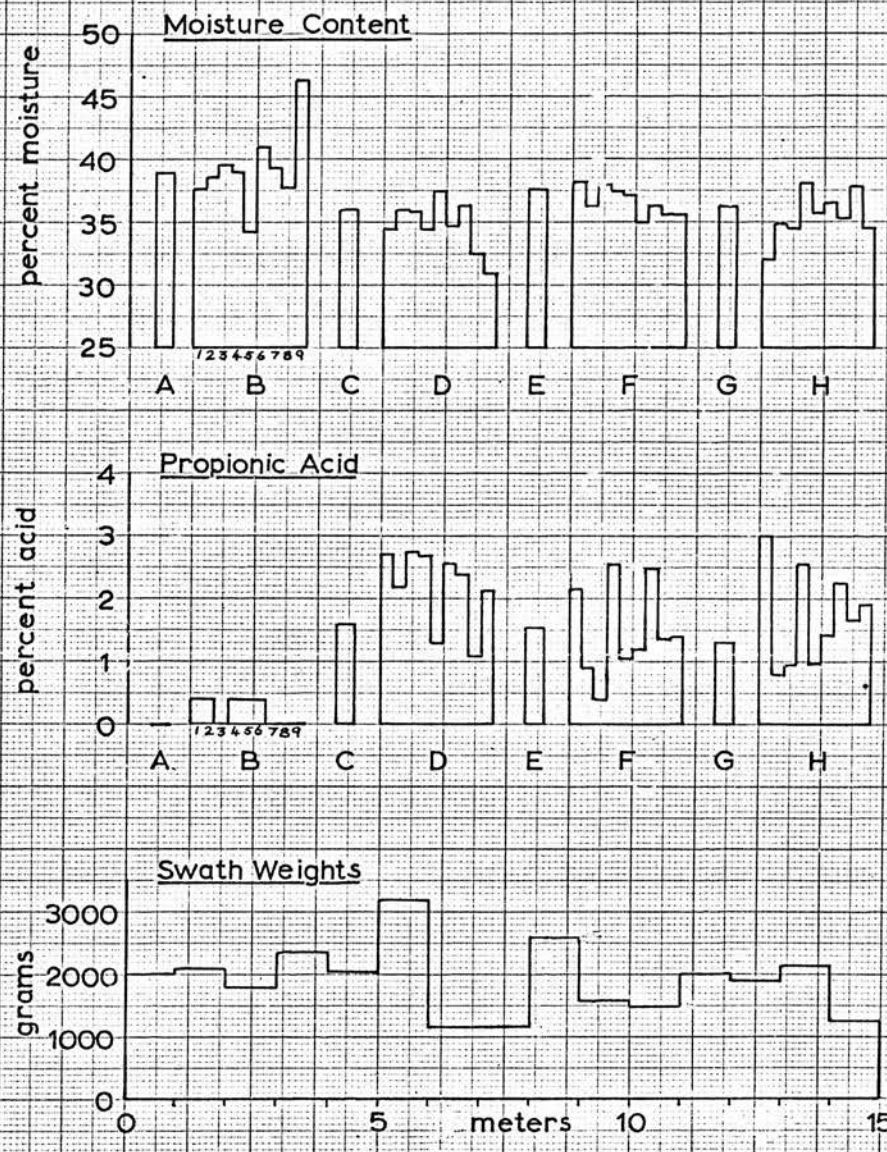
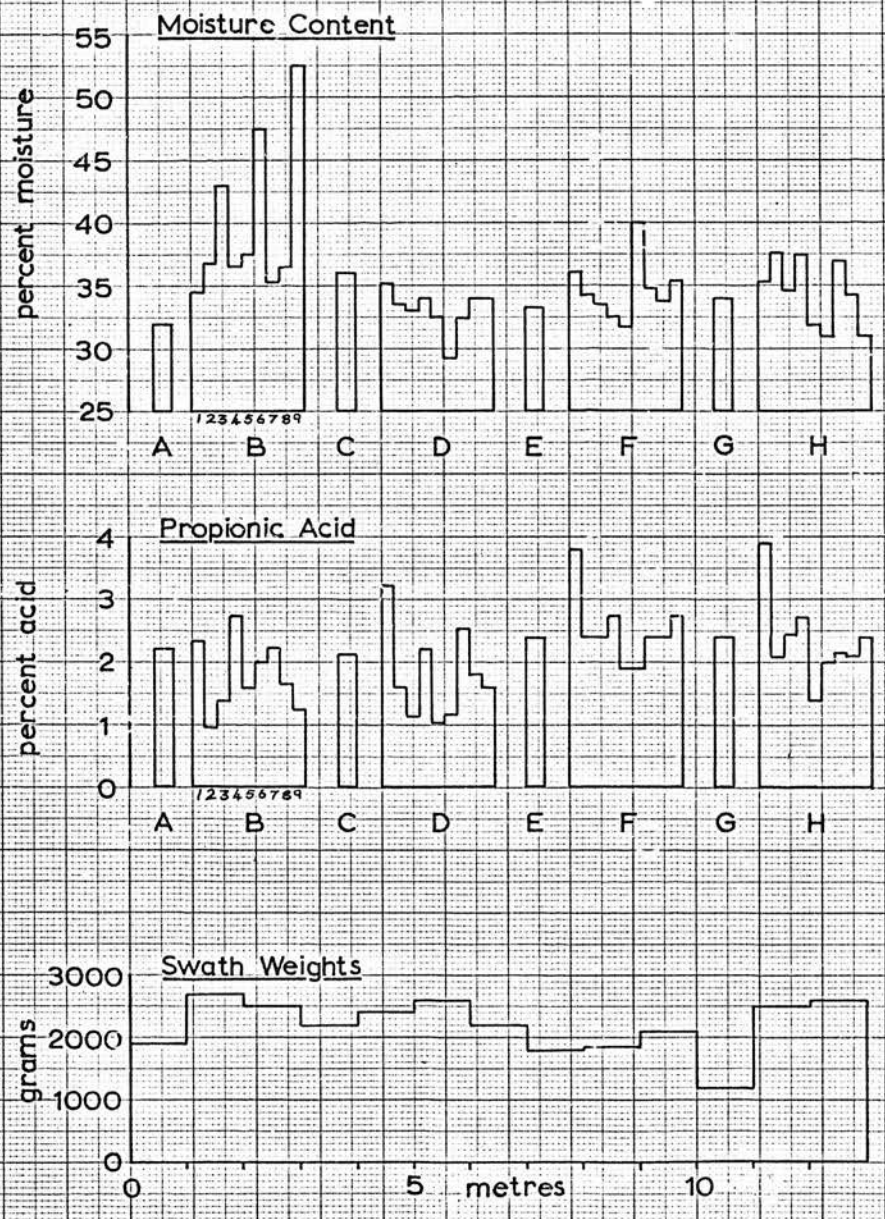


FIGURE 41 1971 BALE ANALYSIS BALE 10



With the propionic acid values a similar trend can be seen, with the coefficients of variation between parts being more than twice those between sections. It is clear even from just looking at the Figures that there are wide variations in both moisture content and acid distribution between the different parts even within a single section of a bale. In section B of bale 10, for example, the moisture content ranged from under 35 per cent to over 52 per cent, while the propionic acid levels in the same section ranged from 1 per cent to 2.7 per cent.

The variations in propionic acid distribution within the section were, however, not entirely random, and it will be seen both in Figure 42 and by studying Figures 40 and 41 that parts 1, 4 and 7 tended to receive significantly more propionic acid than the other parts in the sections.

Figure 42

Mean Propionic Acid Treatments of Parts
(Excluding Section 5B)

	1	4	7	
	2.35%	2.61%	3.04%	
2	1.72%	1.32%	1.56%	8
	1.92%	1.80%	1.64%	
	3	6	9	

Least Significant Difference at P = 0.05

0.59%

Section B of bale 5 received little or no acid, as did section A of the same bale and so it was not included in the calculation of the mean values.

DISCUSSION

The main aim of this experiment was to look at the variations within the bale and to determine the origins and importance of these variations. Although it would seem obvious to look in the swath immediately prior to baling to find the origins of the variations in moisture content and propionic acid distribution it was not possible in this experiment to find any direct link between the swath variations in weight, height and width, and the variations in moisture content and acid distribution found in the bales made from those swaths. There was also found to be very little correlation between the measurements of weight, height and width for each metre length of swath. While it may be true that these various factors are not interrelated, it is more likely to be true that the low correlations were a result of failing to reach the true sources of the variations.

The results in Figure 42 show conclusively that part of the variation in propionic acid distribution within the bales could be attributed to the design of the applicator. The three uppermost parts of each bale section being nearest to the acid spray jets of the applicator, received significantly more acid than the other parts of the bale. However, not all the variation can be attributed to this source and so variations in the swath must be implicated. Measurements on the swath were only taken once every metre and so with a maximum of 15 metres of swath going into any one bale the picture given of the swath at the time of baling is not very detailed, and certainly could not give much information on the minor fluctuations in the swath which might give rise to acid distribution variations.

Another factor enters the picture when we consider the baler. The acid is not sprayed onto the swath as it lies in the field, but onto the swath as it passes through the baler. It is therefore not only possible, but highly likely that variations in the swath were compounded with further irregularities as it was picked up by the baler and before it received the acid treatment.

Thus, concerning the propionic acid distribution in the bales it may be concluded that in bales made from a 'normal' swath and treated with propionic acid from the low pressure applicator fitted to the baler a wide range of acid levels were found in different parts of the bale. Part of this irregular acid distribution could be attributed to the design and position of the acid applicator on the baler and part to the unevenness of the swath at the time of baling and to the way the swath was handled by the baler. Although the swath was shown to be very uneven at the time of baling it was not possible to correlate this directly with variations in acid distribution within the bales made from that swath.

The other major factor under investigation was the moisture content variation found in the bales examined immediately after baling. Once again no direct correlation could be found between the variation in the bales and the variations in swath weights, heights and widths immediately prior to baling. It should be taken into account that these factors were measured after the swaths had been windrowed, i.e., two adjacent swaths had been run together into a single swath which was then of a suitable size for baling. Most of the variations in moisture content, however, would be present in the hay <u>prior</u> to the windrowing and would be most likely due to unevenness in the swaths before they were run together. It is therefore

not surprising that no correlation could be found with the condition of the swath after windrowing. However, the results do clearly show that the moisture content of hay was indeed very variable and although the mean moisture contents of the bales were similar, the values within the bales ranged from 30 per cent to over 50 per cent.

Having established that these variations in moisture content and acid distribution are present in the bales, it was important to make an evaluation of their relative importance. The whole aim of the acid treatment is to preserve the hay, and so with knowledge of the levels of acid required to preserve hay at different moisture contents obtained from the mini-bale experiments it was possible to look at the various parts and sections of the bales in this experiment to find out which parts of the bales received adequate acid treatments to prevent moulding.

In both the propionic acid treated bales of the present experiment the mean level of acid treatment would have been sufficient to preserve the hay at the mean moisture content of the bale if there had been no fluctuations of either factor (Table 25).

Table 25

	Mean Moisture Content	Mean Acid Level	Minimum Adequate Acid Level
Bale 5	36.84%	1.52%	1.31%
Bale 10	34.68%	2.14%	1.18%

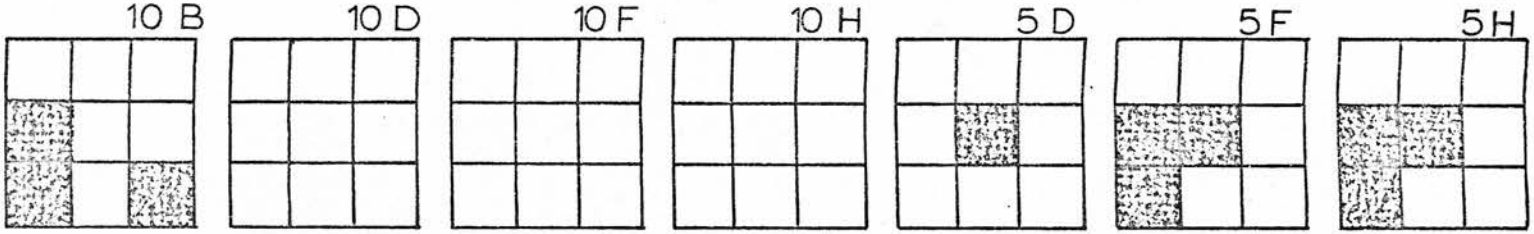
Unfortunately, however, there were fluctuations in both of these factors with the result that several parts of both the propionic acid treated bales received acid levels which would have been inadequate to preserve the hay. In bale 5, discounting Section B which must have been made from hay which entered the baler before the applicator was switched on, 7 out of the remaining 27 individual parts would have been liable to become mouldy, while in bale 10 which received a slightly higher overall acid level, 3 out of the 36 parts were liable to have become mouldy (Figure 43).

It can be seen therefore that variations in moisture content and acid distribution together have resulted in a serious risk of mould patches appearing in both these bales. It does not follow, however, that both factors are equally to blame. If, for example, we consider the hypothetical situation in which the moisture content was to be entirely uniform in both bales, having the same value as the mean of each of the actual bales, but the acid treatments remained exactly as they were in the actual bales: then, as shown in Figure 43, the number of parts in each bale liable to become mouldy would not be reduced. In fact in bale 5 it would rise from 7 to 9 parts, and in bale 10 from 3 to 4 parts. Thus it is obvious that the variations in moisture content have not contributed to the likelyhood of moulding in the bales.

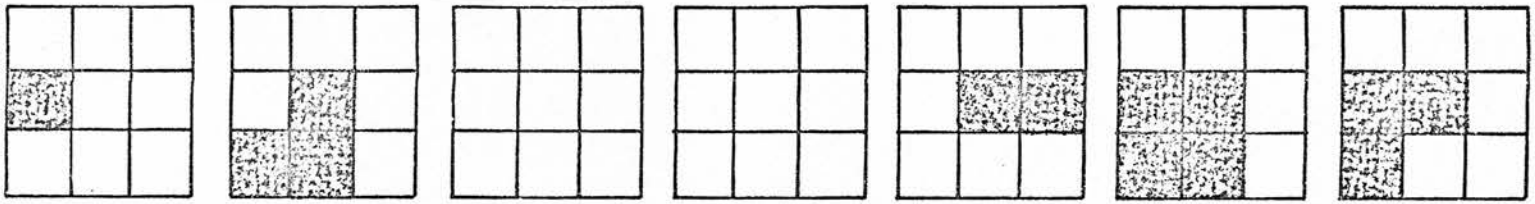
If, on the other hand, the situation is considered in which the moisture content remains variable, but the propionic acid is applied uniformly at the actual mean values, then the number of parts liable to mould would be reduced to only one in the two bales (Figure 43).

FIGURE 43 PARTS OF BALES IN FIRST DISTRIBUTION EXPERIMENT LIABLE TO MOULD

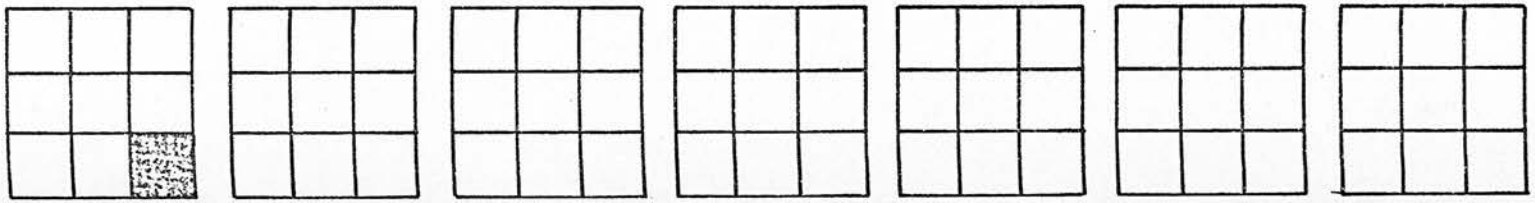
A. Based on the actual moisture content and propionic acid values



B. With a uniform moisture content



C. With a uniform propionic acid distribution



In this experiment, therefore, the results indicate that the variation in propionic acid distribution within the bale and not the variation in moisture content has been the major factor leading to the likelihood of mouldy patches occurring.

Whether this conclusion can be taken to apply more widely than just to the conditions of this experiment is difficult to assess and so it was felt necessary to carry out further experimental work in which the effects of different swath treatments on the acid and moisture variations could be established. This work is described in the Second Distribution Experiment.

SECOND DISTRIBUTION EXPERIMENT

AIMS

The objectives of this experiment were essentially the same as those of the First Distribution experiment. The Batch Experiments described in Section One revealed that a major problem in the propionic acid treatment of hay is the occurrence of patches of mould within the bales. The most likely causes of such mould patches are variations in moisture content and in the distribution of propionic acid. The Distribution experiments were undertaken with the aim of establishing the relative importance of these two factors and to investigate the aspects of the haymaking and treating process which give rise to such variations.

In the first distribution experiment a method was developed by which the pattern of acid and moisture variation within a bale could be measured. The results of the bale analyses were compared with the variations in various parameters which were measured in the swath from which the bales were made. Although yielding some valuable results, the experiment suffered from a number of limitations, which together justified second experiment in which it was hoped that some of the remaining questions would be answered, concerning for example the relationship between the handling and condition of the swath prior to baling and the variations in moisture content within the bale, and also confirm the results of the earlier experiment. The limitations of the first distribution experiment were as follows:

1. Only one swath was used, and this had been handled as it would be under "normal farming practice". "Normal farming practice" is not a precise

term, however, and the experiment gave no indication as to how variations in the handling of the hay in the swath would have affected the final result. This is an important practical consideration, for the methods of handling hay in the field do in fact vary greatly from farm to farm with the result that there will be considerable variation in the state of swaths at the time of baling.

2. The method used for estimating the propionic acid levels on the samples became increasingly inaccurate as the propionic acid level became higher, and levels above about 5 per cent could not be determined with any reasonable degree of accuracy. This was a consequence of using the pH value of the sample as an estimate of the propionic acid content, and may well have resulted in an underestimate of the level of acid on some of the samples. For the present experiment it was considered to be particular importance that the true extent of variation in the acid distribution should be known before any definite conclusions were made.

DESIGN

Four adjacent swaths of hay each 90 metres long were prepared which represented the 'best' and 'worst' of swath conditions in terms of evenness of moisture content and uniformity of 'profile'. By profile is meant the width, height and mass of hay along the length of the swath. Each swath provided a different combination of the two variable factors, profile and moisture content, as follows:-

Swath treatment	A.	Even moisture content; even profile
Swath treatment	B.	Even moisture content; uneven profile
Swath treatment	C.	Uneven moisture content; even profile
Swath treatment	D.	Uneven moisture content; uneven profile

All four swaths were baled and treated with propionic acid in the same way, with the acid being applied at the same rate in each case, and the tractor moving at the same speed. The swaths were baled in quick succession to each other, the whole operation being completed in about half an hour.

From the 8 bales which each swath produced two were selected randomly. These formed two replicates for each of the treatments and were analysed for moisture content and acid distribution in a similar way to the bales in the First Distribution Experiment.

METHODS

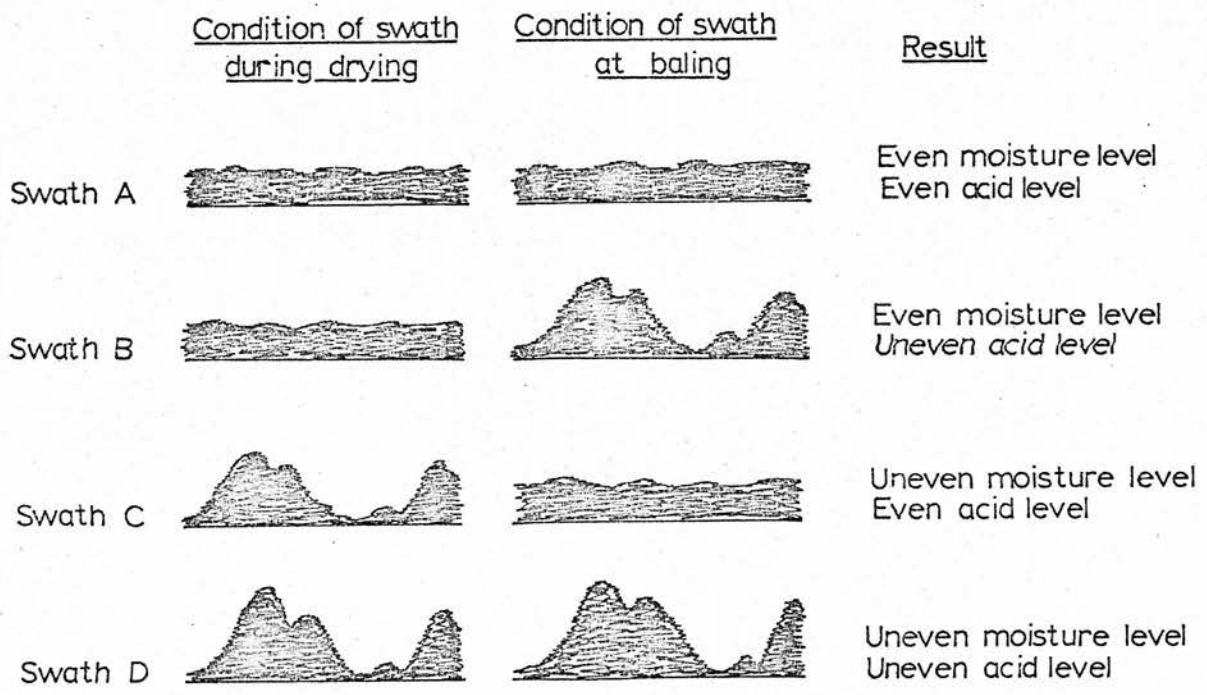
Field Work

Eight adjacent swaths each 90 metres long, were cut in a field of Italian ryegrass using a rotary mower during July 1972. Field treatments were applied to the swaths during the four day drying period as outlined in Table 26.

All the swaths received three teddings and two turnings by means of a tractor powered tedder. Windrowing of the 8 swaths into the 4 treatment swaths was carried out 3 hours before baling. In addition various hand treatments were applied to the swaths in order to achieve the proposed differences between the swaths.

Swath A. This swath was intended to be as even as possible both in moisture content and profile at the time of baling. The two swaths going to make up the final swath were therefore evened out by hand forking on the second day and after windrowing further hand forking was carried out in addition to the machine treatments already mentioned, to achieve the required evenness.

FIGURE 44 Summary of Swath Treatments in the Second Distribution Experiment



Swath B. Up to the time of baling this swath received exactly the same treatment as swath A in order to provide it with as even a moisture content as possible. Immediately prior to baling, however, sections of the swath were gathered into heaps, leaving other parts thin. The swath therefore had a very uneven profile at the time of baling, as can be seen on photograph 11 on page 234.

Swath C. To ensure that this swath had an uneven moisture content, the hay in sections of the swaths before windrowing were gathered into heaps leaving other parts thin with only a covering of hay from the second day of the drying period. The hay was left this way during the machine treatments and up to the time of windrowing. After windrowing and immediately prior to baling however, the uneven profile of the swath was evened out by hand forking so that the swath at baling was uneven in moisture content, but entirely uniform in profile.

Swath D. This swath was treated in exactly the same way as swath C, except that it was not made uniform in profile before baling. It was therefore uneven both in moisture content and profile when it was baled. The treatments are summarised in Figure 44.

During the drying period the moisture contents of the swaths were measured at frequent intervals as in the previous experiments. It was intended to bale at an average moisture content of about 30 per cent but baling in fact took place at about 26 per cent.

The propionic acid was applied using a prototype low pressure applicator supplied by BP Chemicals (International) Ltd. The applicator was of a slightly different design to the one used for the First Distribution

Table 26

Timetable of Operations in the Second Distribution Experiment

1972		
Friday 30th June	Evening	Eight adjacent swaths, each 90 metres long, cut using a rotary mower.
Saturday 1st July	Afternoon	Turned and tedded using tractor
Monday 3rd July	Afternoon	Four of the swaths made very uneven using hand forks. Other swaths evened out.
	Evening	Rain
Tuesday 4th July	All day	Rain
Wednesday 5th July	Afternoon	Weather improved Field turned and tedded using tractor.
Thursday 6th July	Afternoon	Drying weather. Moisture content fell below 40 per cent during afternoon so preparation made to bale that evening.
	3 p.m.	Tractor tedding
	4 p.m.	Windrowing of the 8 swaths into 4 swaths followed by hand treatments as outlined in the Field Work section.
	7 p.m.	Swaths were baled and treated with propionic acid and the bales taken into storage.
Friday 7th July	Morning	Two bales from each treatment cut up using a band saw and samples stored in the deep freeze.

experiment and a full description of it is given in Appendix 7. In this applicator the acid was sprayed from two fan jets situated inside the baler, spraying onto the hay from either side as it passed from the pick-up reel into the bale chamber. In the applicator used previously the two jets both sprayed from above, i.e. down onto the hay as it passed over the pick-up reel. The same tractor speed and acid application rate were used throughout the baling operation. The application rate was set at above the minimum adequate level for two reasons:-

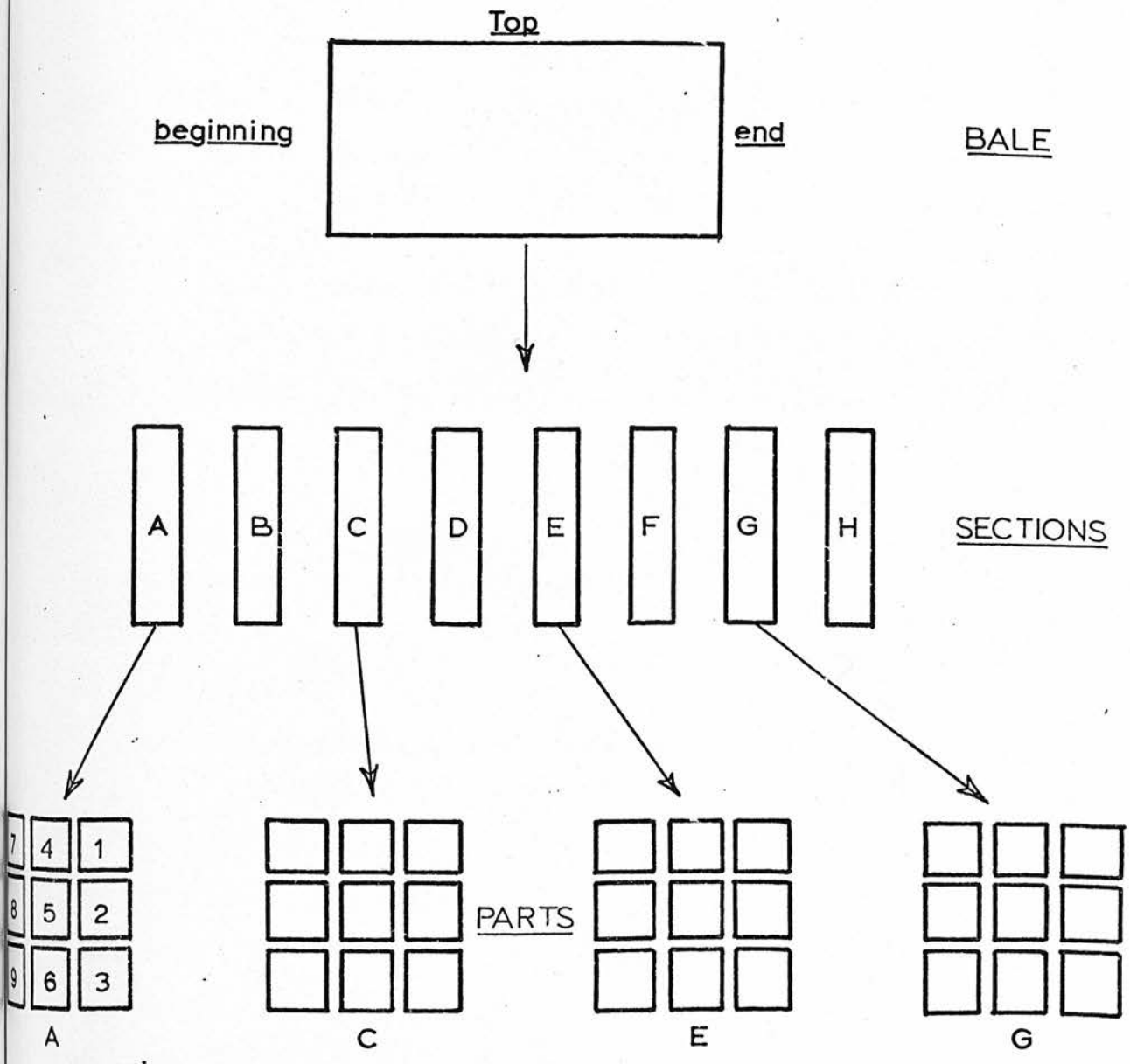
1. Where the application of acid was uneven, a higher rate of acid would tend to give greater differences which would be more easily detected, and less easily confused with inaccuracies in the methods employed for measuring the acid level. The method used in this experiment tended to be less accurate at acid levels below one per cent and so the application of higher acid levels would tend to give more accurate results.

2. Due to the number of samples being handled, there was an inevitable delay of one or two months in the analysis of some samples. Although the samples were to be kept in a deep freeze, the higher level of acid was intended to help to preserve the hay.

The disadvantage in using a high rate of acid application was that it would reduce or eliminate any moulding in bales kept for subsequent examination as few if any parts of the bales would receive an acid level which was inadequate. The effects of variations in the moisture content and acid distribution would therefore be masked by the high overall acid level. The results of this experiment as with the first distribution experiment, however, rest upon the chemical analysis of the bales and not upon the visual assessment of the subsequent moulding, although the latter has provided valuable additional information.

Figure 45

2nd DISTRIBUTION EXPERIMENT
BALE ANALYSIS



Bale Analyses

The 8 bales which had been selected, 2 from each swath, were each divided into 8 equally sized sections. From each bale four of the sections were divided into 9 parts using a band saw as described in the First Distribution experiment. It should be mentioned that the labelling of the parts was slightly different in this experiment as will be seen if Figure 45 is compared with Figure 36 of the First Distribution experiment.

All the portions of the subdivided bales were placed in labelled polythene bags and stored in a deep freeze, from which they were taken in batches as the analyses proceeded. The sectioning and storage of the bales was completed by the afternoon following baling and the analyses took place over a three month period.

Upon removal from the deep freeze each section or part was chopped into centimetre lengths using a Bantall hand powered straw chopper which had been modified by removing one of its two cutting blades. The chopped sample was carefully mixed and subsampled five times for the moisture content and acid level determinations. Two 25 gram subsamples were oven dried in aluminium foil trays and cooled in a desiccator before reweighing to provide duplicate moisture content determinations.

Propionic Acid Determinations

The three remaining sub-samples were used to provide a triplicate propionic acid determination for each part or section. For each determination 15 grams of the chopped sample were placed in a small polythene bag to which 100 ml of deionised water were added. The bag was sealed with a rubber band

and left overnight to soak at room temperature. One corner of the bag was then cut off and the liquid allowed to drain into a beaker. As the hole cut in the bag was small, the remains of the chopped hay sample were not able to escape and were subsequently discarded.

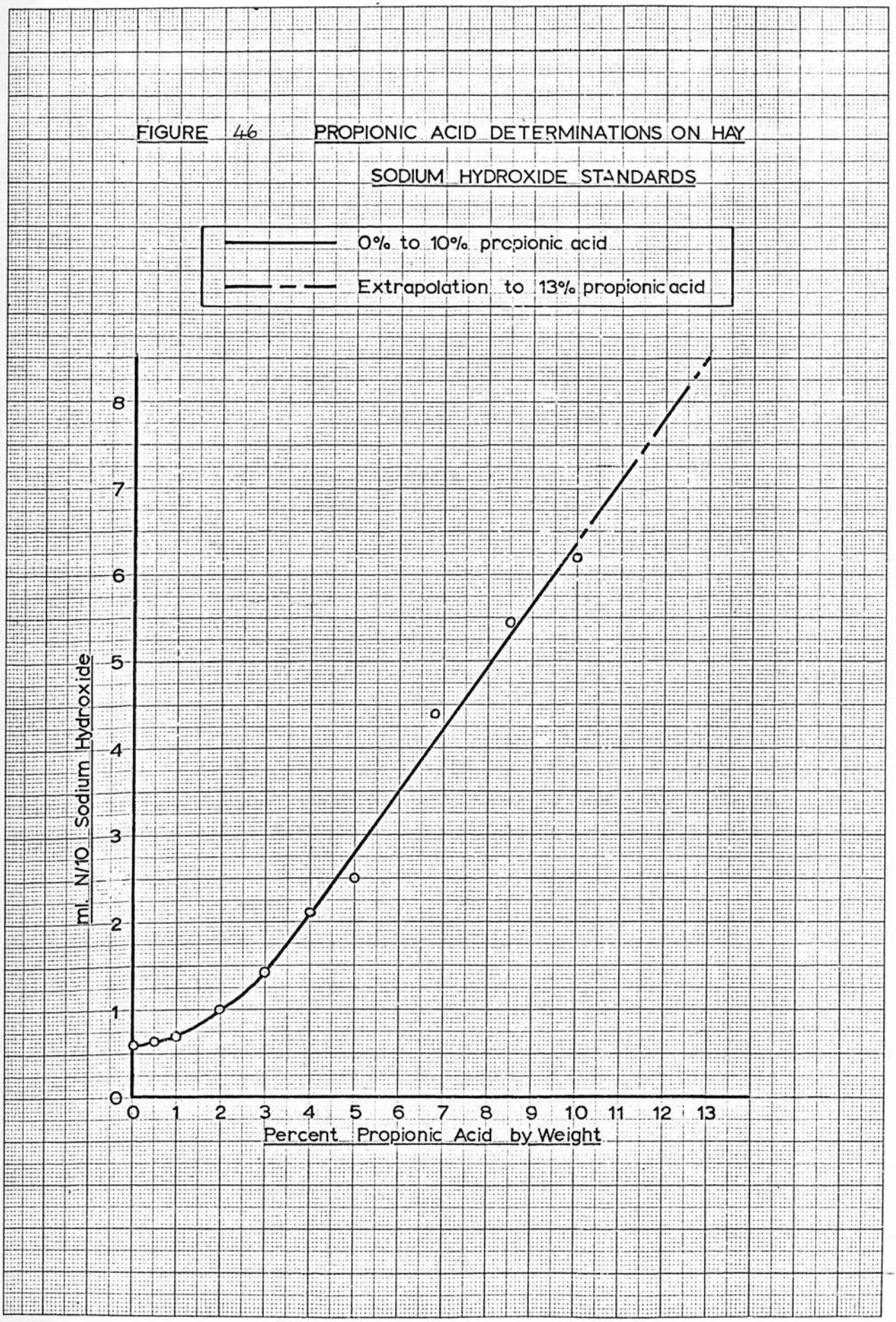
During the period of soaking any acid present would have been able to diffuse out of the hay and into the water, and so by analysing a sample from the resulting solution an estimate of the amount of acid present on the original hay could be obtained. As the propionic acid applied to the hay would be the only source of acidity, the determination was made by neutralising 20 ml. of the liquid with n/10 sodium hydroxide using phenolpthalene indicator. Although some buffering agent from the hay might have been present in the samples, this would not affect the result of the titration.

The recovery of propionic acid using this method was found by calculation to be less than 100 per cent and so a number of standard samples were prepared with hay from the same field as the experiment, treated with propionic acid at known levels ranging from 0.5 per cent to 10.0 per cent by weight. Using these samples, the graph shown on Figure 46 was prepared from which the sodium hydroxide titrations could be translated directly into propionic acid levels on the original hay. A report on the preliminary investigations into this method of propionic acid determination and the results obtained is given in Appendix 10.

The standard samples of treated hay were kept in the deep freeze along with the main samples and analysed periodically with them, to keep a check on any possible changes which might have been taking place in the samples during storage in the deep freeze.

FIGURE 46 PROPIONIC ACID DETERMINATIONS ON HAY

SODIUM HYDROXIDE STANDARDS



Assessment of Visible Moulding

The remaining 24 bales, 6 from each swath, were stored for three months and then an assessment of visible moulding was carried out on 8 of them, 2 being randomly selected from the bales of each swath. Each bale was cut open at 4 places along its length and, using a grid composed of 16 equally sized rectangles, the proportion of mouldy hay was estimated. An outline drawing was then made, using the grid as a guide (Figure 20) of the areas which showed visible moulding together with a brief description of the type of moulding.

RESULTS

The results of this experiment are presented in four sections. Firstly, the effect of the swath treatments on the mean moisture contents of the bales and on the mean acid levels received by the bales are shown in Table 27. Where differences are not significant they are indicated by 'N.S.' The second section of the results concerns the variations in moisture contents and acid levels within the bales as indicated by the coefficients of variation. The variation between the bales' sections (See Figure 45) are considered separately from those between the bales' parts, and the results are presented in Table 28. The results of the bale analyses are also presented in the form of histagrams in Figures 47 to 50.

The third section of the results brings together the moisture contents and acid levels of the parts of the bales to form an estimate of the extent of moulding which would be expected in these bales. Table 31 also shows the extent of moulding which would be expected in certain hypothetical

situations based on the results of the experiment. ~~These results in Table 29 A, calculated on the basis of the propionic acid requirements given in Figure 33, may be compared with the actual extent of moulding found in the bales from the experimental swaths which were stored for three months before being opened. (Table 29 B and Figure 52)~~

Mean Moisture Contents and Propionic Acid Levels

Table 27

A. Mean Bale Moisture Content

per cent	Even moisture Content	Uneven moisture Content	Mean	
Even Profile	23.64	29.50	26.57	N.S.
Uneven Profile	24.13	27.76	25.95	
Mean	23.89	28.63	26.26	

Least Sig. Diff. (P=0.05)
= 2.16

B. Mean Bale Acid Level

per cent	Even moisture Content	Uneven moisture Content	Mean	
Even Profile	3.68	2.46	3.07	N.S.
Uneven Profile	3.69	2.88	3.28	
Mean	3.68	2.67	3.18	

Least Sig. Diff. (P=0.05)
= 0.78

Although an average moisture content of 30 per cent was being aimed at for all the bales, this was not achieved. All the bales had a mean

moisture content below this figure, with an overall mean of 26 per cent. Furthermore, the bales from the 'even moisture' swaths A and B were significantly drier than the bales from the 'uneven moisture' swaths C and D by almost 5 per cent moisture.

The bales from the swaths C and D also received a significantly lower level of acid, by fully 1 per cent, although the setting of the applicator and speed of the baler were the same for all the swaths. As would be expected, the profile of the swath had no effect on the moisture content or acid treatment of the bales.

Moisture Content and Propionic Acid Treatment Variation

Table 28

A. Moisture Content Variation between Sections

Coefficient of Variation	Even moisture Content	Uneven moisture Content	Mean	
Even profile	5.65	5.54	5.60	N.S.
Uneven profile	2.60	6.59	4.60	
Mean	4.13	6.07	5.10	
		N.S.		

B. Moisture Content Variation Between Parts

Coefficient of Variation	Even moisture Content	Uneven moisture Content	Mean
Even profile	7.62	12.99	10.30
Uneven profile	6.74	9.03	7.83
Mean	7.19	11.01	9.09

L.S.D. = 2.29 (P=0.05)

The least variable components of the bales were the moisture contents of the sections, with a mean coefficient of variation of 5.10, compared with 9.09 for the moisture contents of the parts, 29.78 for the acid levels on the sections and 54.88 for the acid levels on the parts. Neither of the swath treatments had any significant effect on the moisture content variation of the sections, but the swath treatment which was intended to make the moisture content more variable did significantly increase the coefficient of variation of the bale parts. The bales made from the swaths with an even profile at the time of baling also showed significantly more variation in the moisture contents of their parts than the bales made from the swaths of uneven profile. The effect of the treatment to give an uneven moisture content was, however, slightly greater than the effect of the uneven profile. In the bales from swath C, which was the most variable, the moisture contents were from 22.03 per cent up to 43.98 per cent, a range of almost 22 per cent, while in swath B which was the least variable the range of 8.5 per cent was from 20.13 to 28.55 per cent.

Table 29

A. Acid Treatment Variation Between Sections

Coefficient of Variation	Even moisture Content		Uneven moisture Content	Mean
Even profile	18.38		16.92	17.64
Uneven profile	45.89		37.53	41.96
Mean	32.11	N.S.	27.44	29.78

L.S.D. = 9.84 (P=0.05)

B. Acid Treatment Variation Between Parts

Coefficient of Variation	Even moisture Content		Uneven moisture Content	Mean
Even profile	47.61		51.17	49.39
Uneven profile	67.46		53.28	60.37
Mean	57.53	N.S.	52.23	54.88

L.S.D. = 10.92 (P=0.05)

FIGURE 47 1972 BALE ANALYSIS
Even Moisture; Even Profile

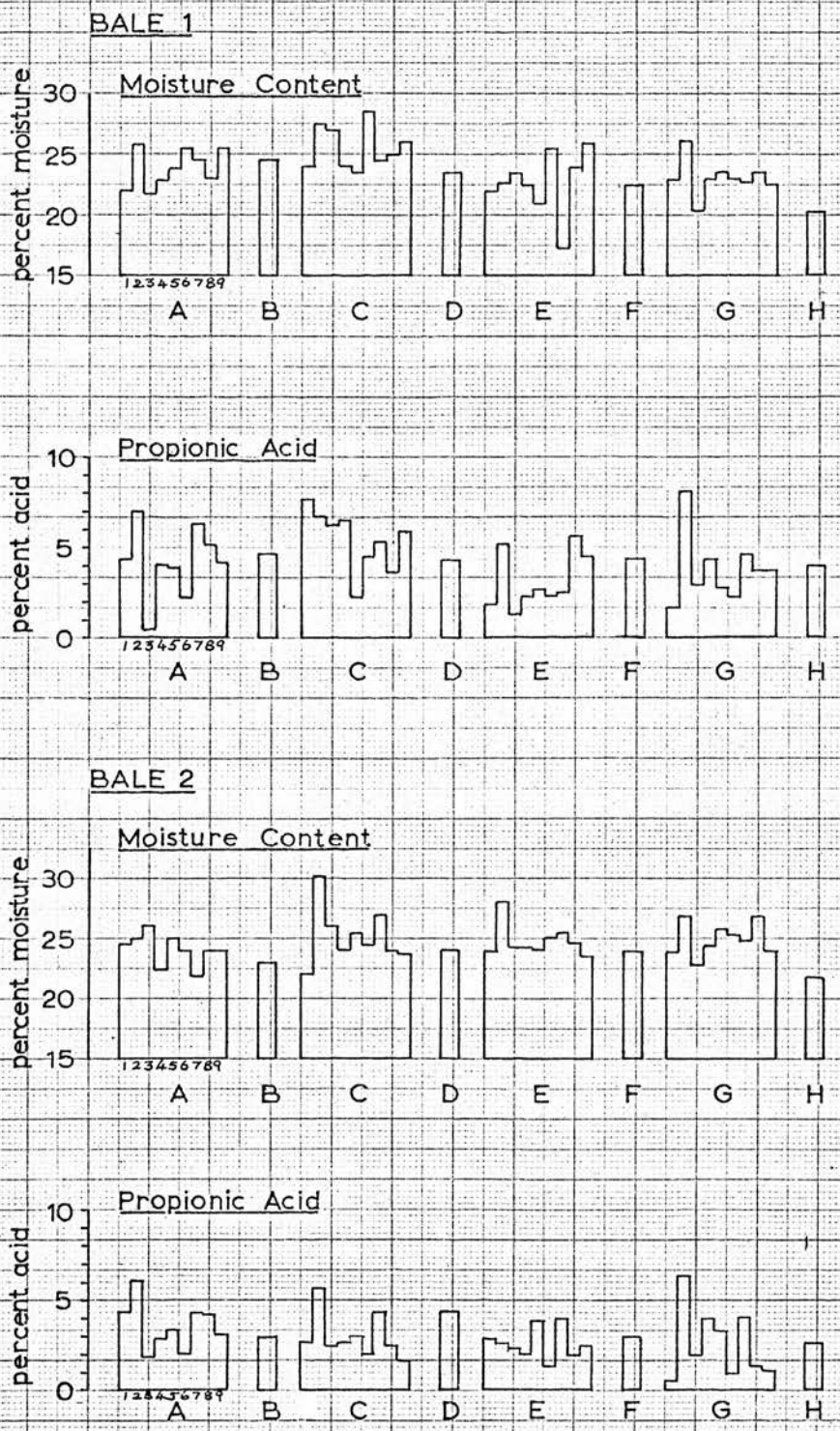


FIGURE 48 1972 BALE ANALYSIS

Even Moisture; Uneven Profile

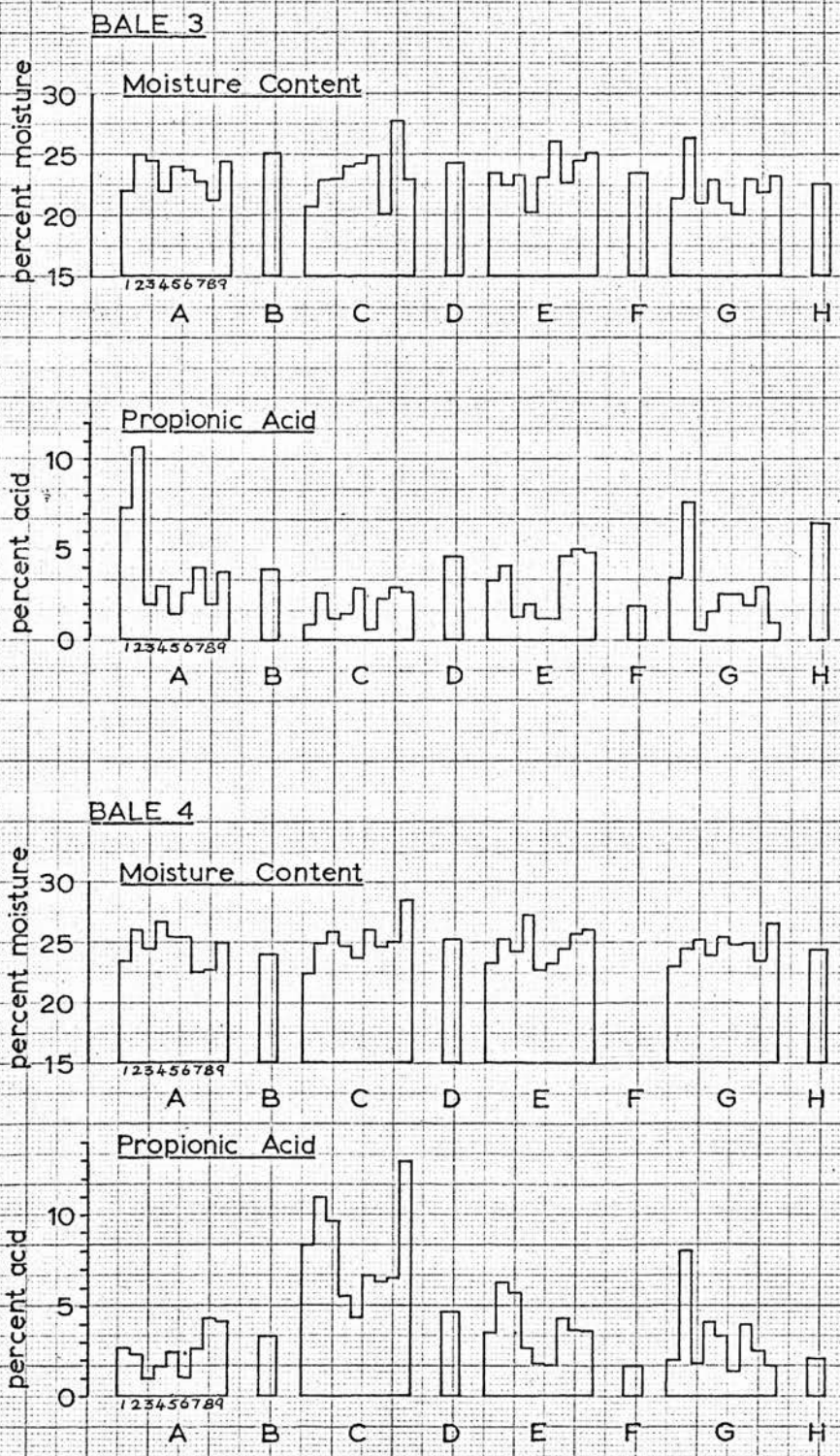


FIGURE 49 1972 BALE ANALYSIS

Uneven Moisture; Even Profile

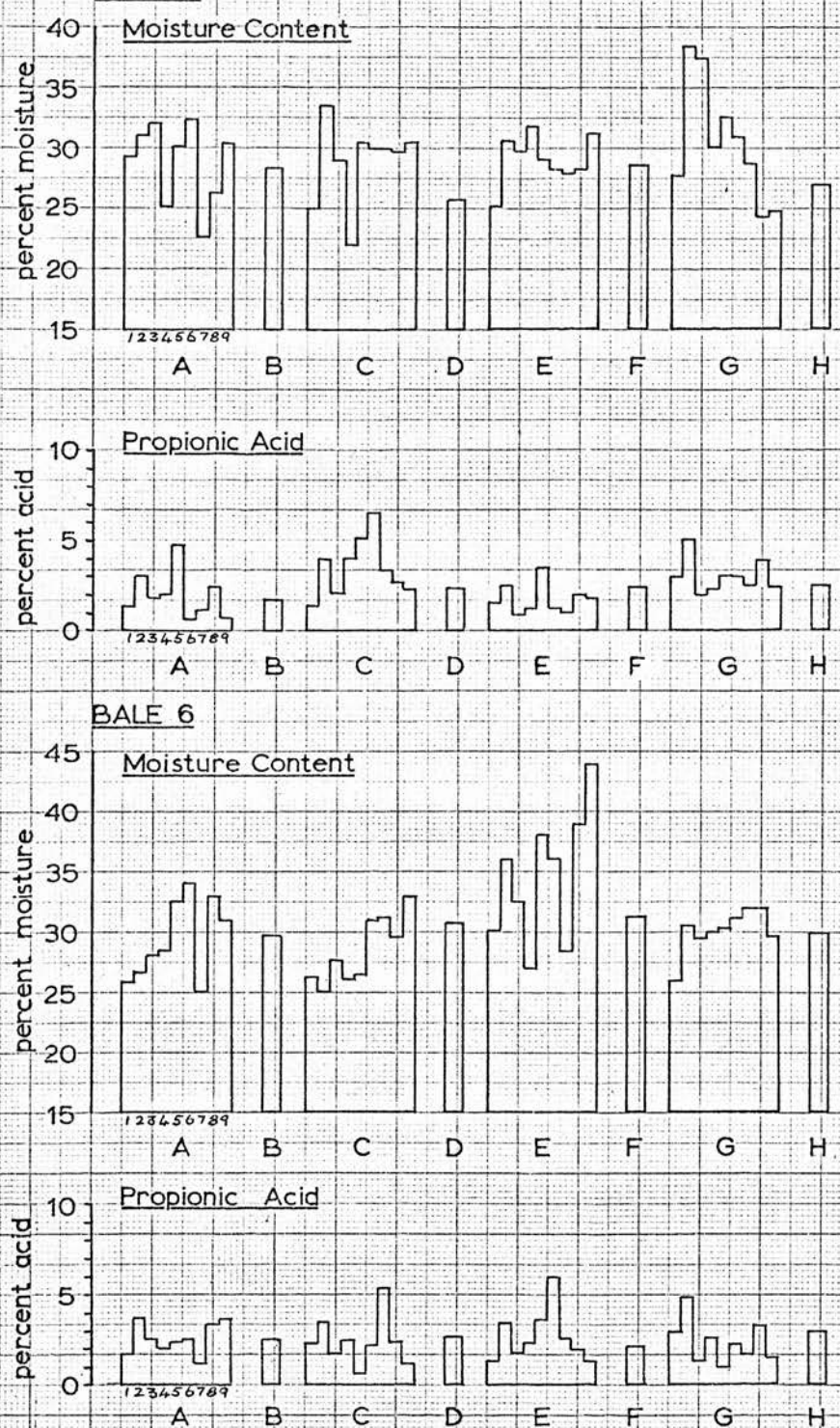
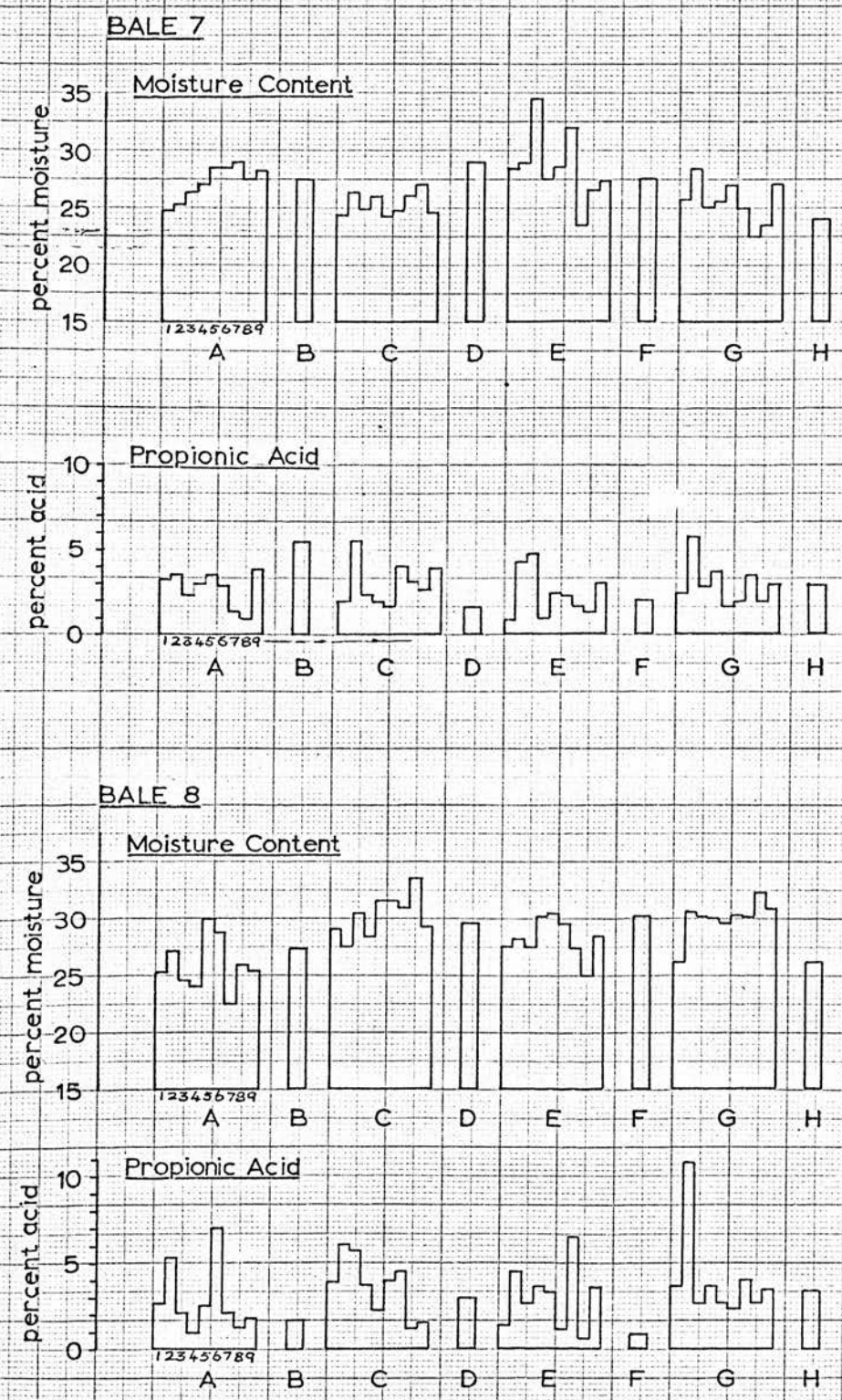


FIGURE 50 1972 BALE ANALYSIS

Uneven Moisture; Uneven Profile



The level of acid on the sections of the bales and on the parts of the bales were very much more variable than the corresponding moisture contents, and by far the most variable was the acid level on the parts of the bales. Over all the bales the treatment levels ranged from 0.5 per cent to 13 per cent on the parts, whereas on the sections the range was from 0.78 to 7.92 per cent.

The swath treatment which made an uneven swath at the time of baling significantly increased the variability of the acid levels both of the sections and of the parts, the effect being most marked on the sections in which the coefficient of variance was raised by over 24 per cent from 17.64 to 41.96 per cent. The moisture content treatments had no effect on the variation in acid levels within the bales.

Figure 51

Mean Propionic Acid Levels on Corresponding Parts of all Bales

	4			
6	3.48	2.89	2.98	1
7	2.94	2.79	5.60	2
8	2.92	2.71	2.61	3
		5		

Least significant difference = 2.33
at P = 0.05

In Figure 51 the tendency for there to be a consistent pattern of acid distribution within the bales is revealed. It can be seen that part 2 . tended to receive a significantly higher level of acid than other

parts of the bales. Although not significantly lower than most of the other parts, part 3 had the lowest mean acid level.

The Consequences of the Acid Level and Moisture Content Variations

The results for each section and part of the bales were studied in the light of the levels of acid required to prevent moulding as shown in Figure 33, and an estimate made of the extent of moulding which would have been expected to take place in these bales. At the end of three months of storage, two of the remaining bales made from each of the experimental swaths were opened and an assessment made of the visible moulding. In Table 30 the estimates from the analysed bales can be compared with the actual values found in the stored bales from the same swaths.

Table 30

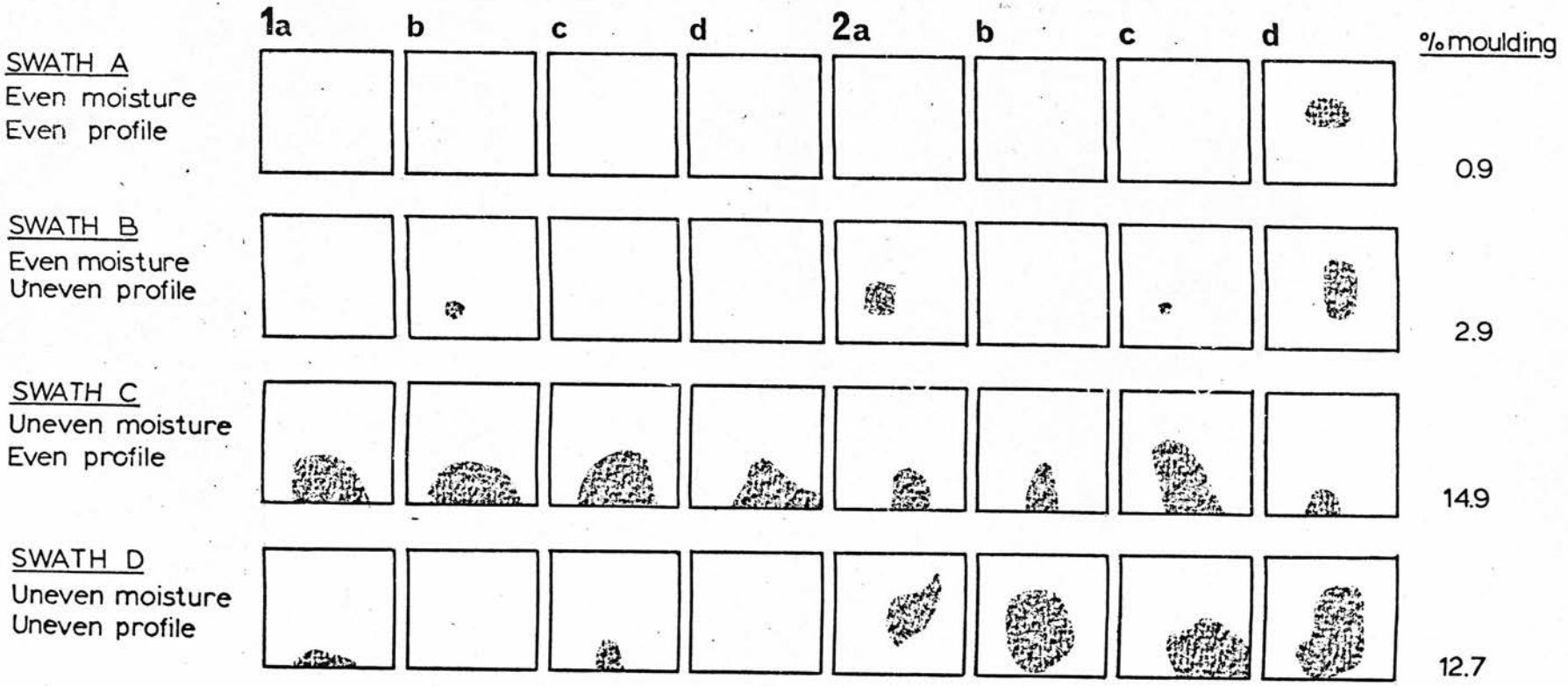
Comparison of Actual Moulding with Predicted Moulding

	Percentage of the hay moulding			
	Even Moisture Even Profile	Even Moisture Uneven Profile	Uneven Moisture Even Profile	Uneven Moist. Uneven Prof.
Actual Moulding	1%	3%	15%	13%
Predicted Moulding	nil	nil	6%	nil

According to the calculations moulding was only expected to take place in the bales from swath C, which received the 'uneven moisture; even profile' treatment, amounting to 6 per cent of the bales. In the actual moulding the bales from this swath did indeed have the most moulding, although it was 2.5 times more extensive than expected. Swaths A and B had very little

FIGURE 52 PATTERN OF MOULDING AFTER THREE MONTHS STORAGE

Two bales selected randomly from each swath



moulding, amounting to not more than 3 per cent of the bales and so could be said to be close to the expected mould free condition, but swath D, which received the 'uneven moisture; uneven profile' treatment contained 13 per cent moulding when none was expected. Thus overall it may be said that the calculated values underestimated the extent of moulding, this being most marked in the swaths which had an uneven moisture content.

In Figure 52 the pattern of moulding in the bales examined at the end of storage is presented. Most of the moulding took place towards the bottom and centre of the bales, with very little at the top and sides of the bales. This pattern corresponds quite closely with that of acid distribution as shown in Figure 51.

In Table 31 the results of this experiment have been used as the basis for constructing three hypothetical situations. In the first case the situation is imagined in which the average moisture content of the hay is known and from this a level of acid 1.5 times the minimum adequate level is calculated from Figure 33. The average moisture content was in fact 26.26 per cent and so 1.5 times the minimum adequate level would be 1.01 per cent acid by weight. One per cent acid is therefore taken to be the mean acid level received by all the bales, corresponding to the 3.18 per cent actually received (Table 27). On the assumption that the pattern of distribution would not be altered by the reduced acid rate, the acid level on each part of the bales is reduced proportionately. From these reduced acid levels the proportion of the bales which would now be expected to mould is calculated, these being the results given in the line A of Table 31.

In the second hypothetical situation, the results for which are shown on line B, the moisture contents were taken to be entirely uniform at the mean

Table 31

Extent of Moulding under Three Sets of Hypothetical Conditions

Percentage Mouldy hay when:-	Swath A Even Moisture Even Profile	Swath B Uneven Moisture Uneven Profile	Swath C Uneven Moisture Even Profile	Swath D Uneven Moisture Uneven Profile
A. All acid levels reduced to give a mean of 1.0%*	13	23	63	34
B. Acid levels as above: Moisture content uniform at 26.26 per cent.+	15	32	42	29
C. Acid level uniform 1.0%*. Moisture contents as in experimental results	nil	nil	26	8

+ 26.26 per cent is the overall mean moisture content of the bales in the experiment.

* 1.0 per cent acid is 1.5 times the minimum acid level required to preserve hay at a moisture content of 26.26 per cent (Figure 33).

value of 26.26 per cent, with the same acid levels being applied at a mean of 1.0 per cent. The situation is reversed in the third set of results for which the acid level was considered to be uniform at 1.0 per cent, but the moisture contents used as they were in the results of the experimental work.

As would be expected, the moulding estimated in the bales at the reduced acid levels (A) was much more extensive than both the estimated moulding in the bales at the full acid levels and the actual moulding in bales from the experimental swaths (Table 30). The results correspond with the observed

moulding in that in the bales from swaths A and B, moulding was estimated as being less severe than in swaths C and D. Swath C, which had the highest moisture content, and the most variable moisture content and received the lowest mean acid level, had the most severe moulding of any of the swaths.

Providing an absolutely uniform moisture content ($\bar{B}$) did not reduce the level of moulding which would be expected in bales from swaths A and B, which already had a relatively even moisture content. In swaths C and D, however, the uniform moisture content did reduce the estimated moulding considerably, the effect being most marked in swath C which, as already stated, had the most variable moisture content.

The hypothetical treatment with a uniform acid level at 1.0 per cent (C), eliminated moulding altogether from the estimates for swaths A and B and considerably reduced the extent of moulding expected in bales from the other swaths. Again swath C had the highest estimate with 26 per cent of the hay moulding. The uniform acid level was more effective in reducing the extent of moulding than the uniform moisture content.

DISCUSSION

So far the work has revealed that although propionic acid can effectively preserve moist hay, and although minimum adequate treatment levels have been established over the most important range of moisture contents, there are still problems concerning the preparation of the swath and the application of the acid. In the Batch experiments, in which the acid was applied to hay as it was baled from the swath, the bales showed moulding in patches within the bales. These mouldy patches may have been a result of poor acid distribution or of variations in the moisture content

of the hay within the bales, or a combination of these two factors.

The First Distribution Experiment indicated that the fluctuation in acid distribution within bales was of greater significance in causing mouldy patches than the variations in moisture content. The fluctuation in acid distribution was attributed in part to the design of the acid applicator and in part to the uneven profile of the swath at the time of baling.

The design of that experiment, however, did not allow for the effect of changes in the condition of the swath at baling upon acid distribution or upon subsequent moulding of the bale. This second distribution experiment was intended to throw some light on this question, and also to provide further evidence in support or otherwise of the conclusions of the first experiment.

While it was intended that all four experimental swaths should have the same average moisture content and acid treatment level, this was not achieved. The swath treatments, in particular the treatment which left the swaths with an uneven profile throughout the drying period resulted not only in making the moisture content more variable, but also in reducing the rate of drying. The bales made from these swaths, C and D, were therefore left with a higher moisture content than the bales from swaths A and B which had an even profile throughout the drying period. From a practical point of view the result could be viewed the other way round, i.e., that the efforts to promote uniform drying of the swath also produced more rapid drying, a conclusion which is not without significance to the farmer.

The extra moisture in the hay of swaths C and D also resulted in these swaths being more bulky. As the acid was applied at the same rate from the applicator for all four swaths it was therefore inevitable that the hay in swaths C and D would receive a lower mean level of acid than the hay in swaths A and B (Table 27B).

Although the swath treatments may have introduced unwanted variations between the bales, they were largely successful in producing the desired effects within the bales. The treatments to produce an uneven moisture content increased the variation in the moisture content of the bale parts, although not of the bale sections, while the treatment to give the swaths an uneven profile increased the variations in acid distribution both on the bale parts and on the bale sections. Thus for both moisture content and acid distribution the major fluctuations occurred in small pockets rather than in large sections of the swath or bale, and the handling of the swath and its condition prior to baling did significantly affect the variations in both of these factors in the bales made from the swath.

In the First Distribution Experiment the coefficient of variation for the moisture contents of the parts had a mean value of 10.82, with a value of 7.46 for that of the sections. These values for what was a 'normal' swath are quite close to the corresponding values for the 'uneven moisture content' swaths, 11.01 and 6.07, in the present experiment. On page 234 a photograph taken during the baling of hay in the First Batch Experiment and showing some typical swaths after windrowing may be compared with a photograph of three of the swaths from the second distribution experiment. In the latter photograph the centre swath has been made particularly uneven, but in fact it could hardly be said to be more uneven than the 'normal' swaths shown in

PLATE 10.

Normal swath about to be baled in the first batch experiment

PLATE 11.

Swaths of even and uneven profile prepared for the second distribution experiment

the first photograph. It is therefore fair to say that none of the swath treatments used in this experiment were outwith the conditions which might be encountered where hay is being made by normal farming methods, and it is the uneven experimental swaths which are more likely to represent the 'typical' swath.

Although the condition of the swath at the time of baling was responsible for much of the variation in acid distribution on the bale parts, the results given in Figure 51 also implicate the design of the propionic acid applicator as being responsible for some of the uneven distribution. It will be remembered that for this second distribution experiment the spray jets were positioned to spray from either side onto the hay as it passed from the pick-up reel of the baler into the bale chamber. This corresponds with pattern of distribution in that the parts of the bale in the middle of one side (part 2) and the top of the other side (part 6) received higher acid levels than the other parts of the bales. In the first distribution experiment a similar result was found, with the top parts of the bales, nearest the spray jets in the earlier design of applicator, receiving the highest acid levels.

The practical significance of these variations in moisture content and propionic acid distribution are seen most clearly both in the moulding which was observed in the bales opened after three months storage and in the estimates of moulding which would be expected to take place in bales under certain conditions. More moulding was found in the bales that had been expected on the basis of the calculated results (Table 30). This divergence could have arisen in a number of ways. It may be that the levels of acid required for preservation of hay shown in Figure 33 and based on the results of the mini-bale experiments are ~~slightly~~ too low. If the

minimum level for adequate preservation were raised by a factor of 100 per cent then the estimates of moulding would correspond more closely. It is also possible that the method used to determine the propionic acid levels on the samples overestimated the amount of acid. This, however, is unlikely as the sodium hydroxide titrations were checked against standard samples of hay treated with known levels of propionic acid at regular intervals.

A third and equally possible reason could be that moulding, having begun in an isolated pocket of the bale, perhaps even smaller than any of the 'parts' in the bale analysis, spread to other areas of the bale which had received a treatment which was marginally adequate. Unfortunately it is not possible from the results of this experiment to find out conclusively which of these factors was responsible for the excess moulding.

The reason for constructing the three hypothetical situations given in Table 31 was to try to gain some information on the relative importance of the variations in acid treatment and moisture content as causes of the subsequent moulding in the bales. The results show that if all variation in acid distribution were to be eliminated, then the propionic acid treatment would be very much more effective in controlling mould growth in the bales. The uniform treatment level of 1.5 times the minimum adequate level of acid was sufficient, in theory, to eliminate all the moulding in swaths A and B which had a relatively uniform moisture content, but to preserve the swaths C and D which had an uneven moisture content, a uniform level of acid at least 2.6 times the minimum adequate level would have been required.

In order to produce a uniform acid distribution improvements would have to be made to the design of the applicator, and a method would have to be

devised to ensure that the swath was entirely even as it passed through the baler.

Creating swaths in which the moisture content was entirely uniform would not reduce moulding as effectively as a uniform acid level, and would only bring the extent of moulding down under conditions in which the moisture content would otherwise have been particularly variable. In practice, as was shown in the swath treatments of this experiment, producing a swath which is uniform in moisture content will also lead to more rapid drying of the swath and would generally be achieved by keeping the swath with an even profile throughout the various swath treatments. Thus, although the provision of swaths with a uniform moisture content is desirable in itself, the success of a propionic acid treatment is more likely to depend upon the evenness of the acid distribution. In the preparation of swaths which are uniform enough in profile to allow this to take place a more even moisture content is likely to occur anyway.

FINAL DISCUSSION

FINAL DISCUSSION

The problems in the use of propionic acid as a preservative for moist hay would not seem to lie in the properties of the chemical itself, for even before this experimental work began it had been established as an effective mould inhibitor (Lewis, 1951). The difficulties lie in the physical and biological properties of the material to be preserved, i.e., the hay. When the propionic preservation of moist grain was introduced relatively few problems were encountered even although, like hay, it is a freshly harvested biologically active material contaminated with many potentially destructive moulds. The success in the treatment of grain lay in the ease with which the chemical could be evenly distributed through the grain, and with the relative homogenity of the grain in terms of moisture content and physical properties.

The nature of hay, however, is entirely different as it has a large surface area and neither good 'flow' characteristics nor usually a uniform moisture content. The variations in moisture content make it difficult for a preservative treatment to be uniformly effective, while the possibility of achieving uniform preservation is made even more remote because it is so difficult to distribute a preservative evenly throughout the hay (Hopkins and Wiant, 1956).

From the previous experimental work summarised in the review, however, propionic acid emerged as the most promising of the chemicals which could be used as a hay preservative. The experimental work of this thesis therefore began with the batch experiments in which the effectiveness of propionic acid was tested under field conditions.

In the first experiment the swaths were prepared using the normal farming procedures of tedding, turning and windrowing and as a result the swaths were not particularly uniform, and even although a fairly sophisticated design of applicator was used, the acid distribution in the bales was found to be uneven. Nevertheless, even at the highest moisture level of hay (44.5 per cent), treated with the lowest level of acid (1.1 per cent), a significant proportion of the hay in the bales was effectively preserved against moulding (Table 10), and the heating reduced, although losses were not. The higher levels of acid at all three moisture levels progressively reduced the losses of dry matter and water soluble carbohydrate and reduced heating and moulding to a greater extent. There is no question, therefore, that propionic acid is capable of reducing moulding and losses in baled hay. The use of 50 bale batches in the second batch experiment provided further support for this conclusion, but also demonstrated even more clearly than in the first experiment that the main failure of the propionic acid treatment was its inability to bring complete and uniform control of moulding except at relatively low moisture contents (Figure 27).

The extent of moulding, however, is not the only way of measuring the success or failure of a treatment, and in the past where different methods of fodder conservation have been compared dry matter loss, rather than moulding, has been used as a basis for comparison. On this criterion a number of propionic acid treatments may be considered to have been successful. When hay has been dried down to the 'safe' 20 per cent moisture level dry matter losses of about 5 per cent can be expected (Watson, Ferguson and Horton, 1937) and in the present experimental work dry matter losses were below 5 per cent in the low moisture hays (below

28 per cent moisture) which had received propionic acid levels of from 2.2 per cent to 5.0 per cent and in the batches of 39.6 per cent moisture which had received 4.1 per cent acid (First Batch Experiment). Thus it may be said that these treatment levels were able to provide effective conservation of the hay dry matter.

Moulding, however, was not entirely prevented in all these batches and so it must be asked if there is an acceptable level of moulding, or whether all moulding must be prevented for hay to be satisfactorily preserved?

Although the organisms which give rise to the most serious condition from mouldy hay, farmer's lung disease, are unlikely to flourish until the temperature in the hay rises to over 40°C, low levels of moulding which do not produce so much heating may still be harmful. Mycotoxins, for example, may be produced in hay which bears only slight mould contamination. It would therefore be dangerous to say that moulding is acceptable at any level, although there must be countless occasions on which farmers have decided to take the risk and have fed mouldy hay rather than waste it altogether.

Thus, although dry matter losses may have been reduced to insignificant levels in the medium moisture hay treated with 4 per cent acid, moulding still took place on 20 per cent of the hay, with water soluble carbohydrate losses of 40 per cent. Although the risk of farmer's lung antigens from this hay was small as the temperature did not rise above 33°C, the level of moulding was such that the acid treatment could not be said to have effectively preserved the hay.

It was only in the low moisture batches which were treated with propionic acid at 2.4, 3.4 and 4.5 per cent in the first experiment, and in the batches receiving 5.0 per cent (but not 2.2 and 3.2 per cent) in the second experiment that no moulding was observed. Very low counts of moulds were found during the microbiological examination of samples from these hays.

On the basis of the two batch experiments it would appear, therefore, that for hay with a moisture content of over about 30 per cent the methods of application of propionic acid which are available at present are unlikely to give adequate protection from moulding even when levels of acid of 5 per cent and over are applied. This conclusion is in line with the finding from the review of previous fieldwork that acid treatments had failed to prevent moulding in any of the hays with a moisture content above 33 per cent.

Also in agreement with earlier findings (A.D.A.S., 1969-1973) is the conclusion that in order to eliminate moulding on hay of under 28 per cent moisture content, levels of propionic acid treatment of 4.5 to 5.0 per cent may be needed when using the types of applicator used in these experiments, but levels of acid of 2.2 per cent and above may bring dry matter losses down to below 5 per cent and prevent moulding in 90 per cent or more of the hay.

During the batch experiments no study was made of the losses of propionic acid which occurred during the treatment process. To have been carried out properly this would have necessitated the analysis of many samples, while the only accurate method of propionic acid determination available at the time was by the use of the silica-gel chromatographic column

technique, which is particularly time consuming and is normally used to separate complex mixtures of fatty acids (Lessard and McDonald, 1966). In connection with the study of the propionic acid distribution, however, a number of samples from one bale were analysed by BP Chemicals (International) Ltd., using the older steam distillation technique. On this one bale, the average level of propionic acid was very close to that which had originally been applied. At that time, therefore, it was not thought that losses of propionic acid during application were significant.

Nevertheless, anyone who was near the front of the baler during the treatment of hay during the batch experiments was well aware of the strong smell of the acid and the danger of catching some of the fine acid spray on the face if there was a gust of wind. Although steps were taken in the second experiment to reduce this spray drift by means of a cover over part of the baler inlet, it is not impossible that losses even as high as the 35 per cent figure reported by Shukking (1972) when using a similar type of applicator, were taking place.

With the Low Pressure Mark II applicator used in the second distribution experiment the jets were much more enclosed, spray drift during operation was minimal, and losses are likely to have been small.

Due to the possibility of significant losses of acid during application in the batch experiments and also to the variations in acid distribution which were known to have taken place, these experiments were valueless in terms of establishing the precise levels of acid application needed to preserve hay at particular moisture contents. In order to make progress in this direction the mini-bale technique was devised in which the quantities of hay would be small enough to be able to be mixed and evenly treated with

propionic acid without difficulty, but large enough to give a reasonable comparison with conventional sized bales. Although in no way connected with this research work, mini-bales have since been proposed as an alternative to conventional bales in commercial systems of haymaking (Anon., 1972).

Some questions may be raised, however, concerning the validity of their use in experiments in which the conditions were meant to be comparable with those in conventional bales.

In neither of the mini-bale experiments were the densities of the bales as high as those in, for example, the bales of the first batch experiment (Appendix 13), although it had been intended that they should be. A number of workers have indicated that visible moulding is more likely to take place in bales which have been made at a higher density (Cashmore and Denham, 1938; Hopkins et al., 1954; Nelson, 1966), and the results of the small experiment reported in Appendix 13 show that higher temperatures will take place in bales of higher density. The evidence from these papers that the density can directly influence moulding is not conclusive, however, and it would seem more likely that where the bale density is higher the increased retention of heat from the initial plant cell respiration leads to the higher temperatures in the bales which in turn stimulates the more rapid growth of moulds. This conclusion is supported by Nelson (1972) who showed that density did not affect dry matter losses and other losses, or the heat output per unit of dry matter. Both Nelson (1972) and Gregory et al., (1963) concluded that moisture content is the most important factor influencing moulding in hay. In the mini-bales the initial plant cell respiration would be virtually eliminated by the death of the cells due to the propionic acid treatment, and so only the subsequent appearance of

moulding, or a change in the ambient temperature could raise the temperature in the bale. While the latter change would be entirely independent of the bale density, the appearance of any moulding at all would indicate that the propionic acid treatment had already failed and so any subsequent effect of the density would not have influenced the main result obtained from the experiment.

Another criticism of the mini-bale technique might arise from the fact that the methods of insulation were unable to prevent a rapid heat loss from the mini-bales. Throughout the experiments the temperature rises, even in the untreated bales, were very much lower than in the equivalent conventional bales. Although the temperatures were lower, this does not necessarily indicate that the actual heat output was any lower. The losses of dry matter and water soluble carbohydrate were, in fact, higher in the mini-bales than in the corresponding untreated conventional bales. Thus, although the actual succession of moulds may have been altered by the lower temperatures, there is no indication that the final extent of the deterioration caused by the moulding was reduced.

It may therefore be reasonably concluded that the slightly lower density of the mini-bales and the lower temperatures in them compared with conventional bales would have had little effect on the outcome of the mini-bale experiments and that the minimum adequate levels of propionic acid treatment suggested in Figure **33** could be taken also to apply to conventional sized bales, providing that the moisture content and acid level within each bale was entirely uniform.

One other point may be raised to question the suggested minimum adequate acid levels, for it is possible that some loss of propionic acid

did take place even during the treatment of the hay for the mini-bales. When standard samples of hay were treated with propionic acid in exactly the same way as for the mini-bales subsequent analyses could not recover 100 per cent of the acid (Appendix 10). Although the possible loss figures of between 15 and 30 per cent which would be needed to explain the recovery results would seem excessively high in view of the very still and cool indoor conditions in which the acid was sprayed onto the hay, this possibility must be acknowledged. The alternative explanation is that the extraction procedure method with water was unable to remove all the acid which may have been converted into propionates in the hay, and this is quite possible for in other methods of analysis hydrochloric acid is used to extract the fatty acids (Lessard and McDonald, 1966).

The conclusions of the mini-bale experiments apply to conditions in which the moisture content and acid distribution in the bales is entirely uniform. The possibility of achieving this condition in conventional sized bales, however, would seem remote and so some thought must be given to the effect of having a range of acid treatments and moisture contents in the bale. The work of Lacey and Hill (1969) and Lacey et al., (1973) at Rothamsted is of particular significance here, for they found that moulding could spread from untreated hay into areas which, under conditions of uniform acid treatment would not have been liable to mould. This spread of moulding could only be prevented by the application of acid levels several times that of the minimum adequate level. Their results also showed that the acid was being degraded and metabolised by the moulds, or at least by the species Paecilomyces varioti, as it spread (Lacey and Lord, 1974)b.

Returning to the present experiments, it may be the case that the minimum levels of propionic acid which were able to preserve hay in the mini-bales under conditions of uniform acid treatment would be unable to be so effective in conventional bales in which the acid treatment is not uniform and mouldy patches appear. Parts of the bales which received acid levels at, or even slightly above, the minimum adequate levels (as estimated from the mini-bale experiments) would be liable to mould due to the influence of adjacent parts of the bales which had moulded as a result of having received a definitely inadequate level.

This may be the reason why, in the second distribution experiment, the moulding observed in the acid treated bales was more extensive than that expected on the basis of the analyses of other bales made from the same swaths (Table 30). The estimates were based on the results of the mini-bale experiments, but, as just described, the moulding could have spread into areas of the bales judged by this standard to be adequately preserved. Only parts of the bales which had received treatments well above the minimum levels would have been safe from the spread of the moulding.

From the results of the second distribution experiment it can be calculated that to bring the predicted figures for moulding in line as nearly as possible with the actual figures the minimum acid levels would have to be almost exactly doubled. If this estimate is reliable, then it would suggest that in order to prevent moulding in a portion of hay which is in contact with mouldy material at least twice the level of acid will be required than would be necessary under conditions of uniform preservation. The equation for the treatment levels could be altered as follows:-

1. Uniform preservation Acid level = (Moisture Content - 15) x 0.06%
2. Variable preservation Acid level = (Moisture Content - 15) x 0.12%

This equation, however, still only applies to portions of the hay with an even acid level and moisture content. If attention is therefore turned from these small parts within the bales to each bale as a whole, it will be recognised that overall levels of acid very much higher than these minimum levels must be applied if preservation is to be achieved. There are three reasons for this. First, as other workers have pointed out (Shukking, 1972; A.D.A.S., 1973), loss of acid is liable to take place during the treatment of the hay and so gross levels of acid application will have to be proportionately higher than the net acid levels required on the hay. Secondly, because the acid is not evenly distributed within the bales the overall level of acid will need to be increased so that even the parts of the bales which tend to receive the lowest levels of acid still receive an adequate treatment. Thirdly, because the variations in moisture content and acid distribution are entirely independent of one another, the acid level applied must be able to preserve hay not at the average moisture content of the hay, but at the maximum moisture content of the hay in the bale.

Together, these factors may account for a considerable escalation of the acid levels which need to be applied, particularly if acid losses are high, the distribution is poor and the moisture content within the bales shows wide variations.

The extent of this escalation may be seen when the actual moisture content and acid distribution results from the second distribution experiment

are used together with the new minimum treatment levels to derive estimates of the levels of acid which would have been necessary if complete, or almost complete preservation of the bales were to have been achieved.

Levels of Acid Required to Preserve Hay in Second Distribution Experiment

	Swath A		Swath B		Swath C		Swath D	
	Even moisture Even profile		Even moisture Uneven profile		Uneven moisture Even profile		Uneven moisture Uneven profile	
Bale	1	2	3	4	5	6	7	8
Minimum adequate acid*	1.0	1.1	1.0	1.2	1.6	1.9	1.4	1.6
Net Acid level to eliminate % moulding	6.4	6.6	6.9	3.6	7.1	6.6	4.9	5.4
Net Acid level to reduce moulding to 3% of hay	4.4	3.6	3.9	4.0	5.9	5.1	4.9	5.3

* At the average moisture content

Net acid treatment levels of between 3.3 and 7 times the minimum adequate levels would have been required in order to eliminate all moulding, although acid levels of between 3 and 4.4 times the minimum would have been sufficient to reduce moulding to less than 3 per cent of the hay. These figures represent actual acid levels of about 7 per cent being required to preserve hay with moisture contents of under 28 per cent moisture, not taking account of losses of acid during the treatment process. If slight moulding on up to 3 per cent of the hay were to be considered acceptable

then the net acid levels could be reduced to about 4.5 per cent on swaths of relatively even moisture content and profile, and 6.0 per cent on swaths of uneven moisture content and profile at mean moisture contents of 24 per cent and 29 per cent respectively.

Supporting evidence which tends to verify the accuracy of these estimates comes from the observed moulding in the bales of the second distribution experiment. In the bales from swaths A and B the actual acid levels applied were 4.2, 3.1, 3.6 and 3.8 per cent respectively for the 4 bales, these values being on average only 0.3 per cent below the levels of acid estimated to bring the moulding to below 3 per cent of the hay. The actual moulding was 1 and 3 per cent for the two swaths. In the bales from swaths C and D the actual acid levels were on average 2.4 per cent below that necessary to reduce moulding below 3 per cent, and the actual moulding was 13 and 8 per cent.

The results of the present investigations therefore indicate that extremely high levels of propionic acid, in the region of 6 or 7 per cent, would be needed to preserve hay made and treated using the techniques currently available, at average moisture contents below 30 per cent. In view of the fact that most of the previous field experiments have used propionic acid levels of between 1 and 4 per cent it is not surprising that on most occasions these treatments have proved to be ineffective. In the second batch experiment, for example, at the relatively low moisture content of 24.8 per cent the 3.2 per cent acid treatment (using the low pressure mark I applicator), was unable to prevent moulding, whereas the 5 per cent acid treatment was effective. Although this 5 per cent figure is below the 6 or 7 per cent acid levels which have been calculated to

prevent all moulding, it is nearer the 4.5 per cent level which would reduce the moulding to less than 3 per cent of the hay, a proportion which could well have remained undetected in that experiment.

In the first batch experiment, however, the propionic acid treatments were effective at even lower levels of application, between 2.4 and 4.5 per cent, on hay of about 27 per cent moisture. In this earlier experiment the high pressure applicator was used which may well have provided a more even distribution of the acid. One of the conclusions from the second distribution experiment was that the most effective way of reducing the levels of acid needed to control moulding would be to provide a better acid distribution, and so the results of the first batch experiment tend to confirm this conclusion.

If it is true, therefore, that a more even distribution of the propionic acid would make this method of hay preservation more likely to be a practical proposition for the farmer, it is important to consider again the results of the second distribution experiment to find out the relative importance of the different factors giving rise to the variations in the treated hay.

From the results of that experiment it is clear that (a) the physical condition of the swath (profile) at the time of baling can significantly affect the distribution of acid; (b) considerable variation in acid distribution can still take place in bales which are made from swaths of relatively uniform profile; (c) a large part of this variation is a result of the positioning of the spray nozzles and the design of the applicator; (d) poor handling of the swath during drying can lead to slower drying and an increase in the moisture content variations; and

(e) variations in moisture content and acid distribution lead to the occurrence of mouldy patches in the bales.

Considering firstly the design of the applicators and the position of the spray nozzles, the patterns given for the low pressure applicators in Figures 42 & 51 clearly indicate that the hay which passed nearest to the spray nozzles received the highest levels of acid. Although the pattern of distribution from the high pressure applicator was also uneven (Table 12), the results were too limited for the effect of the position of the spray bars to be ascertained, although it is likely that the acid distribution was more even than with the low pressure applicators. Once in the bales the pattern of acid distribution is unlikely to change, for, as Lacey and Lord (1972) found, the acid is absorbed by the hay within a few minutes and will not then be picked up in air currents.

The variations in distribution caused by the applicator (coefficient of variation 24 per cent), however, were only responsible for about half of all the variations in acid between the 'parts' of the bales from the swaths with the most even profiles (coefficient of variation, 49 per cent). The remainder of the variation in these swaths must therefore have been caused by minor fluctuations in the profile not evened out by the hand treatments or caused by irregular handling of the hay by the baler; a factor which was not investigated during the experimental work.

In the swaths in which major fluctuations of profile had been introduced artificially the extent of variation in the acid levels was increased, particularly in the bale 'sections' (by 24 per cent), but also between the smaller bale 'parts' (by 11 per cent).

These results are of particular importance for they demonstrate clearly that it is not just the design of the propionic acid applicator which must be improved if even distribution of the propionic acid is to be achieved. Improving the physical condition of the swath at baling together with the way the swath is handled by the baler is at least of equal importance. If it is true that the swaths on most farms resemble the 'uneven' rather than the 'even' swaths in this experimental work, then improving the physical condition of the swath at the time of baling would do more towards an even distribution of acid than re-designing the applicator. As the experimental work also showed, improved handling of the swath to give a more even profile is also likely to bring other benefits in terms of a faster rate of drying and a less variable moisture content, both of which would mean that lower levels of acid application could be used.

Since this experimental work was carried out investigations into a possible method of reducing the variations in acid distribution have begun at the National Institute of Agricultural Engineering at Silsoe. A forward acting tedder has been used to thin out the crop during an acid treatment of the windrow immediately prior to baling (Mandy and Shepperson, 1975). An alternative method of application is also being tried at Newcastle University which would avoid, rather than overcome, the swath variations by applying the acid from the position on the ram face of the baler. The new method of crop conditioning developed by Klinner (1975) may also make propionic acid treatment easier by producing a swath of more uniform profile and moisture content.

A number of practical considerations must also be taken into account if the propionic acid treatment is being advocated for use under normal

farming conditions.

The effective use of propionic acid by the farmer would depend upon his ability to obtain an accurate moisture content for his hay. Purely subjective assessments which have sufficed, rather inadequately, in the past would be of no use when an accuracy of within two per cent is needed. The oven drying method which is standard in laboratories, although providing accuracy, takes 24 hours and therefore could not be used in the context of field work. Even the 'quick drying' method, which was used in this experimental work and took about 30 minutes, often imposed a delay just when time was most critical. Furthermore, both of these methods required the use of an accurate balance for weighing the samples. The 'conductance' type of meters which give an immediate moisture reading for grain samples are less reliable when used with moist hay. At the National Institute for Agricultural Engineering at Silsoe investigations are being carried out into an improved 'conductance' meter which would measure the moisture content of the bales as they leave the baler (Crisford, 1975).

Obtaining a moisture reading from the sample, however, is of little use if the sample itself is not truely representative of the hay to be baled. Whether the farmer, under the pressure of the limited time available at haymaking, would be conscientious enough to spend the time necessary to obtain a representative sample of the field is therefore a vital question.

While propionic acid may be an effective treatment under the right conditions, hay which is intentionally harvested moist, but is inadequately treated with the acid due to a false moisture reading or some other inadvertant error could be liable to much higher losses than if it had been dried completely in the field. Moist hay is also potentially more hazardous

than dry hay due to the risk of the formation of farmer's lung antigens and mycotoxins. The results of the experimental work did show, however, that even where a propionic acid treatment was inadequate to prevent all moulding, it did delay and reduce the heating and moulding of the hay even at moisture contents above 30 per cent. The treatments therefore reduced the risk of farmer's lung antigens forming. This might be taken to be an important point in the favour of propionic treatment of hay, particularly in areas of the country where the incidence of farmer's lung disease due to mouldy hay is high (Grant et al., 1972).

It is therefore possible that the propionic acid treatment of hay, even using the present design of applicators under normal field conditions and at levels of acid significantly below those required to entirely eliminate moulding, could be of use to a farmer who is being forced to bale his hay moist anyway, perhaps due to a complete breakdown in the weather. Under such conditions the acid treatment would provide a low capital cost method of reducing the moulding, the losses and the health hazard of the moist hay. Under better weather conditions the acid could be applied as a safeguard against heating and moulding, if applied at the normal time of baling.

At the present time, however, propionic acid could not be advocated as a reliable method of preserving moist hay which could otherwise be dried to a safe moisture content in the field, or by barn drying, except if extremely high levels of propionic acid are applied to the hay at moisture contents not exceeding 30 per cent. Until such time as the investigations now being carried out to design a more effective method of preparing the swath and providing an even distribution of acid within the bale come to fruition, the full potential of propionic acid as a preservative for moist hay is unlikely to be revealed.

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APPENDICES

APPENDIX 1.

First Batch Experiment

RESULTS AND STATISTICAL ANALYSES

FIRST BATCH EXPERIMENT

Moisture Level High Acid Treatment 1.1 %

BATCH 11*

Initial Moisture Content 44.22 per cent

Initial bale weight	Initial dry matter weight	Final bale weight	Final dry matter per cent	Final dry matter weight	Dry matter Loss per cent
33.792kg	18.849 kg	18.413 kg	76.58	14.101kg	25.19
33.112	18.470	21.962	74.48	16.357	11.44
33.452	18.660	20.236	80.00	16.189	13.24
37.194	20.747	20.965	80.00	16.772	19.16
35.210	19.640	20.418	80.00	16.334	16.83
34.586	19.292	20.349	77.83	15.838	17.90
33.339	18.596	19.321	77.84	15.038	19.13
33.225	18.533	20.568	75.61	15.515	16.28
31.354	17.489	21.022	70.68	14.858	15.04

33.918 mean

Average Dry Matter Loss 17.13 per cent

Average Moisture Loss 68.70 per cent

	Water soluble carbohydrate per cent in D.M.	pH	Moisture content per cent	I.V.O.M. digestibility per cent
Initial	15.46	5.55	44.22	64.6
2 weeks	10.46	5.50	38.60	64.2
6 weeks	6.40	7.14	27.10	62.6
Final	3.00	8.13	21.83	58.4

Water soluble carbohydrate loss 83.92 per cent

* Batches 1 to 10 were not part of this experiment.

FIRST BATCH EXPERIMENT

Moisture Level High Acid Treatment nil

BATCH 12

Initial Moisture Content 44.47 per cent

Initial bale weight	Initial dry matter weight	Final bale weight	Final dry matter per cent	Final dry matter weight	Dry matter Loss per cent
40.256 kg	22.354 kg	20.636 kg	76.47	15.780 kg	29.41
36.401	20.213	21.940	81.01	17.774	12.07
37.875	21.032	21.428	80.00	17.142	18.50
36.627	20.339	21.703	81.08	17.597	13.48
35.493	19.709	20.429	81.08	16.564	15.96
41.844	23.236	25.013	80.00	20.010	13.88
40.143	22.291	22.563	75.33	16.997	23.75
35.040	19.458	21.486	78.42	16.849	13.41
39.916	22.165	23.130	78.71	18.206	17.62

38.177 mean

Average Dry Matter Loss 17.56 per cent

Average Moisture Loss 72.90 per cent

	Water soluble carbohydrate per cent in D.M.	pH	Moisture content per cent	I.V.O.M. digestibility per cent
Initial	19.27	6.06	44.47	66.0
2 weeks	4.10	7.40	23.80	60.2
6 weeks	3.68	8.00	21.30	59.6
Final	2.73	7.94	20.43	56.5

Water soluble carbohydrate loss 88.32 per cent

FIRST BATCH EXPERIMENT

Moisture Level High Acid Treatment 3.2 %

BATCH 13

Initial Moisture Content 41.12 per cent

Initial bale weight	Initial dry matter weight	Final bale weight	Final dry matter per cent	Final dry matter weight	Dry matter Loss per cent
32.658 kg	19.229 kg	23.300 kg	75.23	17.529 kg	8.84
31.751	18.695	25.114	71.74	17.949	3.99
34.133	20.098	22.749	72.46	16.484	17.98
35.720	21.032	26.758	75.42	20.181	4.05
31.694	18.661	25.095	66.01	16.565	11.23
32.545	19.162	28.712	58.99	16.937	11.61
32.431	19.095	20.521	72.00	14.775	22.62
31.751	18.695	23.356	69.28	16.181	13.45
29.880	17.593	23.526	64.31	15.130	14.00

32.507 mean

Average Dry Matter Loss 11.97 per cent

Average Moisture Loss 49.97 per cent

	Water soluble carbohydrate per cent in D.M.	pH	Moisture content per cent	I.V.O.M. digestibility per cent
Initial	18.96	4.90	41.12	57.0
2 weeks	11.53	4.86	37.00	63.2
6 weeks	10.93	4.99	35.90	62.4
Final	6.00	6.19	31.26	57.2

Water soluble carbohydrate loss 72.14 per cent

FIRST BATCH EXPERIMENT

Moisture Level High Acid Treatment 2.1 %

BATCH 15

Initial Moisture Content 45.47 per cent

Initial bale weight	Initial dry matter weight	Final bale weight	Final dry matter per cent	Final dry matter weight	Dry matter Loss per cent
34.586kg	18.860 kg	22.947 kg	77.34	17.747 kg	5.90
32.658	17.808	19.432	73.87	14.354	19.40
30.504	16.634	25.294	62.09	15.705	5.48
36.286	19.787	25.726	68.51	17.625	10.93
33.792	18.427	21.100	73.93	15.599	15.35
30.561	16.665	23.529	71.32	26.781	+0.70
31.978	17.438	22.721	72.32	16.432	5.77
36.514	19.911	22.721	72,03	16.366	17.71
32.885	17.932	21.133	71.27	15,061	16.01

33.307 mean

Average Dry Matter Loss 10.65 per cent

Average Moisture Loss 56.76 per cent

	Water soluble carbohydrate per cent in D.M.	pH	Moisture content per cent	I.V.O.M. digestibility per cent
Initial	13.18	5.18	45.47	62.6
2 weeks	10.52	5.00	35.20	59.9
6 weeks	8.11	5.41	32.30	61.5
Final	4.35	6.93	29.17	59.3

Water soluble carbohydrate loss 70.51 per cent

FIRST BATCH EXPERIMENT

Moisture Level High Acid Treatment 1.1 %

BATCH 16

Initial Moisture Content

Initial bale weight	Initial dry matter weight	Final bale weight	Final dry matter per cent	Final dry matter weight	Dry matter Loss per cent
49.328 kg	27.515 kg	28.564 kg	76.92	21.972 kg	20.15
33.792	18.849	21.827	74.48	16.257	13.75
29.030	16.193	18.030	78.14	14.089	12.99
33.112	18.470	21.931	71.53	15.687	15.07
33.566	18.723	20.823	75.34	15.688	16.21
32.205	17.964	20.354	80.51	16.387	8.78
29.937	16.699	21.420	75.48	16.168	3.18
33.112	18.470	24.879	17.59	17.562	4.92
34.473	19.229	19.673	86.97	17.110	11.12

34.284 mean

Average Dry Matter Loss 11.80 per cent

Average Moisture Loss 65.86 per cent

	Water soluble carbohydrate per cent in D.M.	pH	Moisture content per cent	I.V.O.M. digestibility per cent
Initial	15,46	5.55	44.22	64.6
2 weeks	6.96	6.60	34.90	59.9
6 weeks	2.16	7.45	19.20	60.9
Final	3.31	7.45	25.23	58.7

Water soluble carbohydrate loss 81.12 per cent

FIRST BATCH EXPERIMENT

Moisture Level High Acid Treatment 3.5 %

BATCH 17

Initial Moisture Content 41.64 per cent

Initial bale weight	Initial dry matter weight	Final bale weight	Final dry matter per cent	Final dry matter weight	Dry matter Loss per cent
36.854 kg	21.508 kg	24.427 kg	90.0	21.984 kg	+2.21
28.406	16.578	21.933	72.0	15.792	4.74
31.751	18.530	21.743	69.0	15.003	19.03
35.493	20.714	20.969	72.0	15.098	27.11
34.983	20.416	23.553	67.1	15.802	22.60
38.442	22.435	23.901	71.2	17.010	24.18
32.658	19.059	34.144	90.0	21.730	+14.01
29.710	17.339	25.902	71.0	18.390	+6.06
33.566	19.589	26.639	80.0	21.311	+8.79

33.540 mean

Average Dry Matter Loss 7.40 per cent

Average Moisture Loss 59.35 per cent

	Water soluble carbohydrate per cent in D.M.	pH	Moisture content per cent	I.V.O.M. digestibility per cent
Initial	15.45	4.54	41.64	64.0
2 weeks	11.28	5.01	43.00	60.2
6 weeks	10.13	5.10	35.70	61.9
Final	6.48	5.59	30.32	61.9

Water soluble carbohydrate loss 61.16 per cent

FIRST BATCH EXPERIMENT

Moisture Level High Acid Treatment 2.1 %

BATCH 18

Initial Moisture Content

Initial bale weight	Initial dry matter weight	Final bale weight	Final dry matter per cent	Final dry matter weight	Dry matter Loss per cent
36.117 kg	19.695 kg	21.707kg	78.22	16.979 kg	13.79
32.091	17.499	21.990	81.17	17.849	+2.00
33.792	18.427	22.515	70.69	15.916	13.63
33.792	18.427	25.846	66.77	17.257	6.35
33.735	18.396	23.430	75.49	17.687	3.85
33.205	17.561	24.505	61.33	15.029	14.42
38.328	20.900	20.346	72.49	14.749	29.43
33.566	18.304	21.197	75.42	15.987	12.66
33.000	17.995	24.655	73.31	18.075	+0.44

34.070 mean

Average Dry Matter Loss 10.19 per cent

Average Moisture Loss 59.36 per cent

	Water soluble carbohydrate per cent in D.M.	pH	Moisture content per cent	I.V.O.M. digestibility per cent
Initial	13.18	5.18	45.47	62.6
2 weeks	12.72	4.85	38.70	60.3
6 weeks	4.83	6.81	27.80	59.3
Final	5.43	6.85	27.42	60.2

Water soluble carbohydrate loss 63.00 per cent

FIRST BATCH EXPERIMENT

Moisture Level High Acid Treatment 3.2 %

BATCH 19

Initial Moisture Content 41.12 per cent

Initial bale weight	Initial dry matter weight	Final bale weight	Final dry matter per cent	Final dry matter weight	Dry matter Loss per cent
34.813 kg	20.498 kg	23.410 kg	74.0	17.323 kg	15.49
37.308	21.967	23.240	83.0	19.289	12.19
33.339	19.630	23.930	80.0	19.144	2.48
33.055	19.463	27.152	70.0	19.006	2.35
33.225	19.562	26.148	84.0	21.964	+12.28
34.699	20.431	20.921	91.0	19.038	6.82
36.911	21.733	25.281	87.0	21.994	+1.20
36.060	21.232	21.879	81.0	17.722	12.53
34.813	20.498	28.513	76.0	21.670	+5.72

34.914 mean

Average Dry Matter Loss 3.63 per cent

Average Moisture Loss 66.47 per cent

	Water soluble carbohydrate per cent in D.M.	pH	Moisture content per cent	I.V.O.M. digestibility per cent
Initial	18.96	4.90	41.12	57.0
2 weeks	11.78	4.62	43.30	63.6
6 weeks	6.16	5.48	36.1	61.4
Final	3.07	7.94	25.43	57.9

Water soluble carbohydrate loss 84.60 per cent

FIRST BATCH EXPERIMENT

Moisture Level High Acid Treatment 3.5 %

BATCH 20

Initial Moisture Content 41.64 per cent

Initial bale weight	Initial dry matter weight	Final bale weight	Final dry matter per cent	Final dry matter weight	Dry matter Loss per cent
37.081 kg	21.640 kg	22.494 kg	71.00	15.971 kg	26.20
30.958	18.067	24.367	70.00	17.057	5.59
33.452	19.523	18.131	88.00	15.955	18.28
32.999	19.258	26.408	68.49	18.087	6.08
35.493	20.714	24.144	82.00	19.798	4.42
32.999	19.258	25.906	72.00	18.652	3.15
33.679	19.655	27.145	—	—	—
34.586	20.184	24.310	82.00	19.934	1.24
35.097	20.483	19.094	88.00	16.803	17.97

34.038 mean

Average Dry Matter Loss 10.37 per cent

Average Moisture Loss 62.48 per cent

	Water soluble carbohydrate per cent in D.M.	pH	Moisture content per cent	I.V.O.M. digestibility per cent
Initial	15.45	4.54	41.64	64.0
2 weeks	9.64	5.00	40.50	57.6
6 weeks	5.06	7.00	32.4	58.5
Final	4.92	7.06	29.70	57.7

Water soluble carbohydrate loss 71.46 per cent

FIRST BATCH EXPERIMENT

Moisture Level Medium Acid Treatment 4.1 %

BATCH 21

Initial Moisture Content 39.56 per cent

Initial bale weight	Initial dry matter weight	Final bale weight	Final dry matter per cent	Final dry matter weight	Dry matter Loss per cent
29.370 kg	17.751 kg	23.418 kg	71.01	16.629 kg	6.32
30.164	18.231	27.897	70.73	19.732	+8.34
28.803	17.409	21.686	73.44	15.926	8.52
30.617	18.505	24.042	78.85	18.957	+2.44
30.164	18.231	27.159	69.91	18.987	+4.15
30.617	18.505	26.571	63.66	16.915	8.59
29.370	17.751	22.738	67.34	15.312	13.74
29.134	17.614	22.794	69.94	15.942	9.49
30.731	18.574	29.315	63.99	18.759	+1.00

29.886 mean

Average Dry Matter Loss 3.41 per cent

Average Moisture Loss 35.66 per cent

	Water soluble carbohydrate per cent in D.M.	pH	Moisture content per cent	I.V.O.M. digestibility per cent
Initial	16.64	4.68	39.56	62.9
2 weeks	12.32	4.79	34.70	62.2
6 weeks	12.10	4.80	33.90	59.0
Final	9.56	5.10	31.32	62.3

Water soluble carbohydrate loss 44.51 per cent

FIRST BATCH EXPERIMENT

Moisture Level Medium Acid Treatment 3.3 %

BATCH 22

Initial Moisture Content 33.48 per cent

Initial bale weight	Initial dry matter weight	Final bale weight	Final dry matter per cent	Final dry matter weight	Dry matter Loss per cent
27.442 kg	18.354 kg	23.645 kg	73.56	17.393 kg	4.72
30.277	20.140	24.155	73.22	17.686	12.03
26.875	17.877	25.130	69.40	17.440	2.44
26.989	17.953	25.734	69.01	17.765	1.15
26.762	17.802	24.367	70.34	17.140	3.72
29.427	19.575	26.891	64.56	17.361	11.31
31.751	21.121	25.038	73.67	18.479	12.51
29.824	19.839	27.387	71.09	19.469	1.87
28.973	19.273	27.954	69.30	19.372	+0.51

28.702 mean

Average Dry Matter Loss 5.47 per cent

Average Moisture Loss 21.14 per cent

	Water soluble carbohydrate per cent in D.M.	pH	Moisture content per cent	I.V.O.M. digestibility per cent
Initial	12.15	4.61	33.48	62.7
2 weeks	14.13	4.58	34.30	61.5
6 weeks	11.45	4.77	32.60	59.1
Final	12.02	4.97	31.25	64.9

Water soluble carbohydrate loss 6.48 per cent

FIRST BATCH EXPERIMENT

Moisture Level Medium Acid Treatment 1.0 %

BATCH 23

Initial Moisture Content 36.05 per cent

Initial bale weight	Initial dry matter weight	Final bale weight	Final dry matter per cent	Final dry matter weight	Dry matter Loss per cent
29.597 kg	18.927 kg	22.210 kg	73.33	16.287 kg	13.95
30.958	19.798	21.700	82.14	17.824	9.97
30.731	19.652	21.985	84.07	18.483	5.95
32.772	20.958	23.287	83.89	19.535	6.79
29.143	18.637	21.934	79.80	17.503	6.08
26.875	17.187	21.249	81.56	17.331	+0.84
33.225	21.247	23.684	82.70	19.587	7.81
30.164	19.290	21.020	82.42	17.325	10.19
26.308	16.824	22.210	73.54	16.333	2.92

29.975 mean

Average Dry Matter Loss 6.98 per cent

Average Moisture Loss 59.83 per cent

	Water soluble carbohydrate per cent in D.M.	pH	Moisture content per cent	I.V.O.M. digestibility per cent
Initial	15.27	5.48	36.05	63.2
2 weeks	6.34	6.55	20.90	59.9
6 weeks	6.90	5.95	18.50	60.6
Final	5.64	6.60	20.21	62.6

Water soluble carbohydrate loss 65.64 per cent

FIRST BATCH EXPERIMENT

Moisture Level Medium Acid Treatment nil

BATCH 24

Initial Moisture Content 34.59 per cent

Initial bale weight	Initial dry matter weight	Final bale weight	Final dry matter per cent	Final dry matter weight	Dry matter Loss per cent
29.937 kg	19.582 kg	21.652 kg	82.80	17.928 kg	8.45
33.792	22.103	23.240	81.65	18.975	14.15
32.205	21.065	22.901	82.29	18.845	10.54
32.829	21.473	23.410	84.74	19.837	7.62
31.071	20.324	21.339	83.03	17.718	12.82
32.658	21.362	19.692	83.86	16.514	22.69
36.854	24.106	26.075	81.89	21.353	11.42
34.019	22.252	21.822	82.52	18.008	19.07
33.792	22.103	23.410	81.52	19.084	13.66

33.017 mean

Average Dry Matter Loss 13.38 per cent

Average Moisture Loss 65.74 per cent

	Water soluble carbohydrate per cent in D.M.	pH	Moisture content per cent	I.V.O.M. digestibility per cent
Initial	19.43	6.34	34.59	63.8
2 weeks	5.38	6.95	20.6	59.8
6 weeks	5.36	6.46	17.70	59.4
Final	3.52	6.91	18.18	57.7

Water soluble carbohydrate loss 84.30 per cent

FIRST BATCH EXPERIMENT

Moisture Level Medium Acid Treatment 3.3 %

BATCH 25

Initial Moisture Content 33.48 per cent

Initial bale weight	Initial dry matter weight	Final bale weight	Final dry matter per cent	Final dry matter weight	Dry matter Loss per cent
28.916 kg	19.235 kg	25.509 kg	73.59	18.772 kg	2.41
30.164	20.065	20.633	67.77	13.983	30.31
28.916	19.235	21.895	73.28	16.045	16.58
25.514	16.972	23.978	68.83	16.504	2.76
28.066	18.670	15.889	69.25	17.928	3.97
29.597	19.688	25.525	74.14	18.924	3.88
29.257	19.462	26.756	65.71	17.581	9.66
29.937	19.914	20.803	75.31	15.667	21.33
30.107	20.027	22.164	75.10	16.645	16.89

28.942 mean

Average Dry Matter Loss 11.98 per cent

Average Moisture Loss 29.93 per cent

	Water soluble carbohydrate per cent in D.M.	pH	Moisture content per cent	I.V.O.M. digestibility per cent
Initial	12.15	4.61	33.48	62.7
2 weeks	12.61	4.81	33.40	61.9
6 weeks	10.46	4.91	32.10	62.1
Final	11.18	5.21	29.87	59.3

Water soluble carbohydrate loss 18.99 per cent

FIRST BATCH EXPERIMENT

Moisture Level Medium Acid Treatment 2.3 per cent

BATCH 26

Initial Moisture Content 34.29 per cent

Initial bale weight	Initial dry matter weight	Final bale weight	Final dry matter per cent	Final dry matter weight	Dry matter Loss per cent
29.937 kg	19.672 kg	24.534 kg	71.61	17.569kg	10.69
27.896	18.330	19.885	81.98	16.302	11.06
28.123	18.480	19.953	78.36	15.635	15.40
31.184	20.491	22.153	78.84	17.465	14.77
25.514	16.765	19.320	73.56	14.212	15.23
32.885	21.609	21.466	—	—	—
28.803	18.926	20.905	73.96	15.461	18.31
27.102	17.809	23.230	71.83	16.686	6.31
27.669	18.181	21.812	76.05	16.588	8.76

28.790 mean

Average Dry Matter Loss 12.57 per cent

Average Moisture Loss 44.71 per cent

	Water soluble carbohydrate per cent in D.M.	pH	Moisture content per cent	I.V.O.M. digestibility per cent
Initial	14.24	4.91	34.29	64.8
2 weeks	7.22	5.90	31.80	60.5
6 weeks	7.31	6.30	25.60	60.9
Final	7.42	6.24	25.48	65.6

Water soluble carbohydrate loss 54.44 per cent

FIRST BATCH EXPERIMENT

Moisture Level Medium Acid Treatment 4.1%

BATCH 27

Initial Moisture Content 39.56 per cent

Initial bale weight	Initial dry matter weight	Final bale weight	Final dry matter per cent	Final dry matter weight	Dry matter Loss per cent
28.803 kg	17.409 kg	24.997 kg	72.26	18.063 kg	+3.76
29.143	17.614	23.183	74.62	17.299	1.79
28.690	17.340	22.118	79.29	17.537	+1.14
28.803	17.409	21.255	73.36	15.593	10.43
30.334	18.334	24.243	69.03	16.735	8.72
29.370	17.751	25.381	67.09	17.028	4.07
28.009	16.929	24.941	69.79	17.406	+2.82
29.483	17.820	27.776	66.27	18.407	+3.29
27.896	16.860	24.374	67.61	16.479	2.26

28.948 mean

Average Dry Matter Loss 1.81 per cent

Average Moisture Loss 38.18 per cent

	Water soluble carbohydrate per cent in D.M.	pH	Moisture content per cent	I.V.O.M. digestibility per cent
Initial	16.64	4.68	39.56	62.9
2 weeks	11.52	4.79	37.70	63.3
6 weeks	6.23	5.62	28.63	62.7
Final	10.90	5.04	28.87	60.9

Water soluble carbohydrate loss 35.69 per cent

FIRST BATCH EXPERIMENT

Moisture Level Medium Acid Treatment 1.0%

BATCH 28

Initial Moisture Content 36.05 per cent

Initial bale weight	Initial dry matter weight	Final bale weight	Final dry matter per cent	Final dry matter weight	Dry matter Loss per cent
30.107 kg	19.253 kg	20.568 kg	82.43	16.954 kg	11.94
36.287	23.206	23.913	79.57	19.028	18.00
27.442	17.549	20.862	74.21	15.482	11.78
31.184	19.942	22.496	81.08	18.240	8.53
33.509	21.429	23.923	82.81	19.811	7.55
26.932	17.223	23.700	70.13	16.621	3.50
31.289	20.015	21.362	79.00	16.876	15.68
31.014	19.833	21.589	79.44	17.150	13.53
29.200	18.673	22.893	77.73	17.795	4.70

30.775 mean

Average Dry Matter Loss 10.58 per cent

Average Moisture Loss 56.59 per cent

	Water soluble carbohydrate per cent in D.M.	pH	Moisture content per cent	I.V.O.M. digestibility per cent
Initial	15.27	5.48	36.05	63.2
2 weeks	16.01	4.81	31.30	63.1
6 weeks	5.72	6.22	21.80	59.6
Final	6.94	6.60	21.10	62.6

Water soluble carbohydrate loss 59.36 per cent

FIRST BATCH EXPERIMENT

Moisture Level Medium Acid Treatment nil

BATCH 29

Initial Moisture Content 34.59 per cent

Initial bale weight	Initial dry matter weight	Final bale weight	Final dry matter per cent	Final dry matter weight	Dry matter Loss per cent
34.246 kg	22.400 kg	22.042 kg	83.15	18.328 kg	18.18
34.473	22.549	21.475	81.95	17.599	21.95
34.700	22.697	22.508	78.15	17.590	22.50
28.973	18.951	20.795	84.68	17.609	7.08
31.978	20.917	22.798	84.49	19.262	7.91
33.225	21.732	22.222	83.02	18.449	15.11
46.974	30.708	32.021	82.15	26.305	14.34
30.220	19.767	21.192	82.88	17.564	11.14
30.107	19.693	21.135	81.78	17.284	12.23

33.874 mean

Average Dry Matter Loss 14.49 per cent

Average Moisture Loss 65.67 per cent

	Water soluble carbohydrate per cent in D.M.	pH	Moisture content per cent	I.V.O.M. digestibility per cent
Initial	19.43	6.34	34.59	63.8
2 weeks	8.82	6.39	20.20	60.2
6 weeks	3.50	6.66	16.70	60.4
Final	3.88	7.09	16.50	59.6

Water soluble carbohydrate loss 82.92 per cent

FIRST BATCH EXPERIMENT

Moisture Level Medium Acid Treatment 2.3 %.

BATCH 30

Initial Moisture Content 34.29 per cent

Initial bale weight	Initial dry matter weight	Final bale weight	Final dry matter per cent	Final dry matter weight	Dry matter Loss per cent
27.839 kg	18.293 kg	19.503 kg	75.87	14.797 kg	19.11
25.231	16.579	19.729	77.35	15.260	7.96
30.447	20.007	19.807	81.92	16.226	18.90
26.989	17.734	23.131	69.28	16.025	9.64
28.349	18.628	26.656	65.44	17.444	6.36
28.349	18.628	21.513	74.75	16.081	13.67
28.009	18.405	26.080	66.10	17.239	6.34
28.803	18.926	21.430	76.15	16.319	13.77
28.236	18.554	22.678	69,97	15.868	14.48

28.028 mean

Average Dry Matter Loss 12.25 per cent

Average Moisture Loss 36.10 per cent

	Water soluble carbohydrate per cent in D.M.	pH	Moisture content per cent	I.V.O.M. digestibility per cent
Initial	14.24	4.91	34.29	64.8
2 weeks	13.14	4.82	31.90	61.1
6 weeks	11.73	5.00	30.50	61.1
Final	6.74	5.95	26.18	65.5

Water soluble carbohydrate loss 58.47 per cent

FIRST BATCH EXPERIMENT

Moisture Level Low Acid Treatment 1.0 %

BATCH 31

Initial Moisture Content 26.53 per cent

Initial bale weight	Initial dry matter weight	Final bale weight	Final dry matter per cent	Final dry matter weight	Dry matter Loss per cent
26.535 kg	19.495 kg	22.061 kg	81.50	17.980kg	7.77
29.653	21.786	25.293	81.38	20.583	5.52
27.499	20.204	24.239	81.49	19.752	2.24
23.757	17.454	21.664	79.46	17.214	1.38
28.236	20.745	21.717	77.13	16.750	19.26
25.514	18.745	22.481	81.98	18.430	1.68
30.617	22.494	27.958	76.39	21.375	5.05
29.427	21.620	25.350	76.76	19.459	9.83
24.267	17.829	21.834	75.73	16.535	7.26

27.278 mean

Average Dry Matter Loss 6.67 per cent

Average Moisture Loss 31.62 per cent

	Water soluble carbohydrate per cent in D.M.	pH	Moisture content per cent	I.V.O.M. digestibility per cent
Initial	16.05	5.63	26.53	61.7
2 weeks	10.64	5.18	27.00	66.2
6 weeks	14.08	5.80	24.90	64.2
Final	10.73	5.79	23.61	64.7

Water soluble carbohydrate loss 37.61 per cent

FIRST BATCH EXPERIMENT

Moisture Level low

Acid Treatment 3.4 %

BATCH 32

Initial Moisture Content 27.48 per cent

Initial bale weight	Initial dry matter weight	Final bale weight	Final dry matter per cent	Final dry matter weight	Dry matter Loss per cent
28.009 kg	20.312 kg	26.361 kg	77.46	20.419 kg	+0.53
24.607	17.845	23.470	76.90	18.048	+1.14
28.463	20.641	25.198	77.43	19.511	5.47
24.721	17.928	23.810	76.76	18.277	+1.95
23.813	17.269	23.014	77.17	17.760	+2.84
23.360	16.941	22.363	77.92	17.425	+2.86
24.380	17.680	23.980	75.35	18.069	+2.20
24.040	17.434	22.959	75.17	17.258	1.01
25.458	18.462	23.753	74.90	17.791	3.63

25.206 mean

Average Dry Matter Loss +0.16 per cent

Average Moisture Loss 19.23 per cent

	Water soluble carbohydrate per cent in D.M.	pH	Moisture content per cent	I.V.O.M. digestibility per cent
Initial	12.26	4.64	27.48	63.1
2 weeks	7.73	5.02	27.40	63.5
6 weeks	12.24	5.18	25.00	61.0
Final	13.16	4.91	27.44	68.2

Water soluble carbohydrate loss +7.51 per cent

FIRST BATCH EXPERIMENT

Moisture Level Low Acid Treatment 4.5 %.

BATCH 33

Initial Moisture Content 26.44 per cent

Initial bale weight	Initial dry matter weight	Final bale weight	Final dry matter per cent	Final dry matter weight	Dry matter Loss per cent
27.272 kg	20.061 kg	25.970 kg	77.26	20.064 kg	+0.01
28.973	21.313	28.182	76.83	21.652	+1.59
28.123	20.687	26.563	74.93	19.904	3.78
27.442	20.186	26.367	76.90	20.276	+0.45
26.195	19.269	22.558	77.69	17.526	9.05
25.968	19.102	24.689	75.76	18.705	2.08
26.875	19.769	25.120	75.59	18.988	3.95
24.267	17.851	23.872	74.90	17.880	+0.16
25.798	18.977	25.573	75.58	19.328	+1.74

26.768 mean

Average Dry Matter Loss 1.66 per cent

Average Moisture Loss 14.32 per cent

	Water soluble carbohydrate per cent in D.M.	pH	Moisture content per cent	I.V.O.M. digestibility per cent
Initial	16.41	4.59	26.44	62.9
2 weeks	13.47	4.54	28.00	62.4
6 weeks	17.36	4.70	27.70	62.8
Final	17.43	4.66	29.19	70.2

Water soluble carbohydrate loss +4.46 per cent

FIRST BATCH EXPERIMENT

Moisture Level Low Acid Treatment 2.4 % .

BATCH 34

Initial Moisture Content 26.82 per cent

Initial bale weight	Initial dry matter weight	Final bale weight	Final dry matter per cent	Final dry matter weight	Dry matter Loss per cent
24.494 kg	17.925 kg	24.999kg	73.61	18.402 kg	+2.66
25.855	18.921	24.773	72.50	17.960	5.08
28.349	20.746	26.470	73.29	19.400	6.49
23.870	17.468	22.732	71.58	16.272	6.85
24.494	17.925	23.404	75.68	17.712	1.19
23.870	17.468	22.701	72.82	16.531	5.36
23.984	17.551	22.902	71.84	16.453	6.26
24.947	18.256	24.603	70.69	17.392	4.73
23.984	17.551	23.525	72.38	17.027	2.99

24.872 mean

Average Dry Matter Loss 4.03 per cent

Average Moisture Loss 1.78 per cent

	Water soluble carbohydrate per cent in D.M.	pH	Moisture content per cent	I.V.O.M. digestibility per cent
Initial	13.24	4.86	26.82	61.1
2 weeks	14.80	4.98	26.20	62.4
6 weeks	16.31	5.10	25.30	62.3
Final	13.66	4.84	27.82	65.1

Water soluble carbohydrate loss 0.99 per cent

FIRST BATCH EXPERIMENT

Moisture Level Low Acid Treatment nil

BATCH 35

Initial Moisture Content 26.48 per cent

Initial bale weight	Initial dry matter weight	Final bale weight	Final dry matter per cent	Final dry matter weight	Dry matter Loss per cent
27.556 kg	20.259 kg	22.052 kg	84.65	18.667 kg	7.86
26.308	19.342	20.011	84.42	16.893	12.66
26.705	19.634	21.329	83.96	17.908	8.79
25.741	18.925	20.774	85.84	17.832	5.78
29.937	22.010	23.102	85.27	19.699	10.50
26.195	19.259	21.159	85.78	18.150	5.76
26.819	19.717	20.407	82.88	16.913	4.22
26.989	19.842	21.258	81.86	17.402	2.30
27.499	20.217	22.052	83.00	18.303	9.47

27.083 mean

Average Dry Matter Loss 7.48 per cent

Average Moisture Loss 55.68 per cent

	Water soluble carbohydrate per cent in D.M.	pH	Moisture content per cent	I.V.O.M. digestibility per cent
Initial	13.99	6.33	26.48	65.3
2 weeks	9.04	5.85	20.40	61.7
6 weeks	8.51	6.00	15.60	62.2
Final	6.42	5.86	15.73	62.4

Water soluble carbohydrate loss 57.54 per cent

FIRST BATCH EXPERIMENT

Moisture Level Low Acid Treatment 1.0 %

BATCH 36

Initial Moisture Content 26.53 per cent

Initial bale weight	Initial dry matter weight	Final bale weight	Final dry matter per cent	Final dry matter weight	Dry matter Loss per cent
26.989 kg	19.829 kg	22.958 kg	75.27	17.280 kg	12.85
24.834	18.246	22.448	79.39	17.821	2.33
23.417	17.204	19.971	79.41	15.859	7.82
26.648	19.578	21.598	83.83	18.106	7.52
29.030	21.328	24.337	80.82	19.669	7.78
26.252	19.287	22.580	76.89	17.362	9.98
22.623	16.621	19.897	74.71	14.865	10.56
26.422	19.412	22.448	77.44	17.384	10.45
24.550	18.037	21.257	81.62	17.350	3.81

25.641 mean

Average Dry Matter Loss 8.12 per cent

Average Moisture Loss 31,73 per cent

	Water soluble carbohydrate per cent in D.M.	pH	Moisture content per cent	I.V.O.M. digestibility per cent
Initial	16.05	5.63	26.53	61.7
2 weeks	14.76	5.50	27.90	62.8
6 weeks	12.62	5.90	22.30	61.9
Final	9.78	5.73	22.35	62.4

Water soluble carbohydrate loss 44.02 per cent

FIRST BATCH EXPERIMENT

Moisture Level Low Acid Treatment 4.5 %.

BATCH 37

Initial Moisture Content 26.44 per cent

Initial bale weight	Initial dry matter weight	Final bale weight	Final dry matter per cent	Final dry matter weight	Dry matter Loss per cent
26.025 kg	19.144 kg	24.594 kg	71.34	17.545 kg	8.35
26.989	19.853	25.784	73.18	18.869	4.96
27.272	20.061	26.081	73.53	19.177	4.41
27.102	19.936	26.238	71.65	18.800	5.70
25.741	18.935	24.847	71.43	17.748	6.27
29.597	21.771	28.181	69.98	19.721	9.42
25.401	18.685	25.444	71.84	18.279	2.17
25.004	18.393	24.650	72.18	17.792	3.27
25.911	19.060	25.671	70.27	18.039	5.36

26.560 mean

Average Dry Matter Loss 5.55 per cent

Average Moisture Loss +3.67 per cent

	Water soluble carbohydrate per cent in D.M.	pH	Moisture content per cent	I.V.O.M. digestibility per cent
Initial	16.41	4.59	26.44	62.9
2 weeks	12.89	5.08	27.10	62.2
6 weeks	14.64	4.95	27.10	63.2
Final	13.71	4.50	29.03	62.5

Water soluble carbohydrate loss 21.09 per cent

FIRST BATCH EXPERIMENT

Moisture Level Low Acid Treatment nil

BATCH 38

Initial Moisture Content 26.48 per cent

Initial bale weight	Initial dry matter weight	Final bale weight	Final dry matter per cent	Final dry matter weight	Dry matter Loss per cent
28.349 kg	20.842 kg	22.157 kg	84.69	18.765 kg	9.97
24.664	18.133	19.718	84.91	16.743	7.67
29.483	21.676	23.346	83.93	19.510	9.99
27.669	20.342	22.610	86.36	19.526	4.01
25.344	18.633	22.476	85.33	19.179	+2.93
24.097	17.716	19.092	84.10	16.068	9.30
26.648	19.592	21.476	82.85	17.793	9.18
29.257	21.510	23.971	82.69	19.822	7.85
27.839	20.467	21.930	82.72	18.140	11.37

27.039 mean

Average Dry Matter Loss 7.38 per cent

Average Moisture Loss 51.69 per cent

	Water soluble carbohydrate per cent in D.M.	pH	Moisture content per cent	I.V.O.M. digestibility per cent
Initial	13.99	6.33	26.48	65.3
2 weeks	10.72	5.89	25.60	61.2
6 weeks	8.87	5.92	16.10	64.4
Final	6.60	5.91	15.72	59.2

Water soluble carbohydrate loss 56.30 per cent

FIRST BATCH EXPERIMENT

Moisture Level Low Acid Treatment 3.4%

BATCH 39

Initial Moisture Content 27.48 per cent

Initial bale weight	Initial dry matter weight	Final bale weight	Final dry matter per cent	Final dry matter weight	Dry matter Loss per cent
23.927 kg	17.352 kg	21.824 kg	71.52	15.609kg	10.04
26.422	19.161	24.659	78.00	19.234	+0.38
25.514	18.503	24.608	75.00	18.456	0.25
25.628	18.585	24.376	76.45	18.635	+0.27
25.514	18.503	24.472	73.36	17.953	2.97
24.891	18.051	23.871	73.52	17.550	2.88
25.968	18.832	25.226	72.90	18.390	2.35
23.870	17.311	23.979	71.01	17.027	1.64
25.401	18.421	25.283	72.44	18.315	0.58

25.237 mean

Average Dry Matter Loss 2.23 per cent

Average Moisture Loss 8.47 per cent

	Water soluble carbohydrate per cent in D.M.	pH	Moisture content per cent	I.V.O.M. digestibility per cent
Initial	12.26	4.64	27.48	63.1
2 weeks	15.00	4.83	26.00	74.6
6 weeks	17.87	4.86	26.10	64.7
Final	13.85	4.81	26.81	61.7

Water soluble carbohydrate loss +10.45 per cent

FIRST BATCH EXPERIMENT

Moisture Level Low Acid Treatment 2.4%.

BATCH 40

Initial Moisture Content 26.82 per cent

Initial bale weight	Initial dry matter weight	Final bale weight	Final dry matter per cent	Final dry matter weight	Dry matter Loss per cent
24.891 kg	18.215 kg	22.005 kg	74.03	16.290 kg	10.57
24.607	18.007	23.820	74.31	17.701	1.70
27.442	20.082	26.226	75.20	19.722	1.80
25.174	18.422	23.650	74.84	17.700	3.92
22.226	16.265	21.889	72.95	15.968	1.83
26.138	19.128	22.826	74.10	16.914	11.57
25.855	18.921	24.387	72.29	17.629	6.83
23.473	17.178	23.480	71.63	16.819	2.09
26.762	19.584	25.407	71.65	18.204	7.05

25.174 mean

Average Dry Matter Loss 5.26 per cent

Average Moisture Loss 6.62 per cent

	Water soluble carbohydrate per cent in D.M.	pH	Moisture content per cent	I.V.O.M. digestibility per cent
Initial	13.24	4.86	26.82	61.1
2 weeks	11.56	4.81	25.50	62.5
6 weeks	15.98	4.90	26.50	62.4
Final	14.97	4.85	27.03	63.9

Water soluble carbohydrate loss +7.12 per cent

FIRST BATCH EXPERIMENT

ANALYSIS OF VARIANCE

Percentage Water Soluble Carbohydrate Losses

Moisture	Block	nil	1%	2%	3%	4%	Total
43	A	88.32	83.92	70.51	72.14	61.16	376.05
43	B	-	81.12	63.00	84.60	71.46	300.18
36	A	84.30	65.64	54.44	6.48	44.51	255.37
36	B	82.92	59.36	58.47	18.99	35.69	255.43
26	A	57.54	37.61	0.99	+7.51	+4.46	84.17
26	B	56.30	44.02	+7.12	+10.45	21.09	103.84
	Total	369.38	371.67	240.29	164.25	229.45	1375.04

Treatment Totals

Moisture	nil	1%	2%	3%	4%	Total
43	88.32	165.04	133.51	156.74	132.62	676.23
36	167.22	125.00	112.91	25.47	80.20	510.80
26	113.84	81.63	+6.13	+17.96	16.63	188.01
Total	369.38	371.67	240.29	164.25	229.45	1375.04

Total of replicate A's 715.59 Total of replicate B's 659.45

Correction for the mean = 65197.8

Analysis of Variance Table

	df	Sum of Sqs.	Mean Sqs.	Variance Ratio
A vs B	1	2.6		
Moisture	2	15238.3		
Moisture vs block	2	36.2		
Moisture plots	5	15277.1		
Acid level	4	8007.7	2001.9	36.5
Moisture vs acid	8	4805.3	600.7	11.0
Error	11	603.2	54.8	
Total	28	28693.3		

Standard Error of acid treatment means $= \sqrt{\frac{54.8}{6}} = 3.022$

Least significant difference between means $= 3.022 \times 2.2 \times \sqrt{2}$

$= 9.4$ (P = 0.05)

FIRST BATCH EXPERIMENT

Analysis of Variance (cont.)

Analyses of the remaining results were carried out in exactly the same way as for the water soluble carbohydrate losses. The following values for the standard error and least significant differences were found:-

	Standard Error	Least significant difference (p=0.05)
Mean initial bale weight	0.28	0.88 kg
Final moisture content	0.68	2.11%
Final W.S.C. content	0.49	1.53%
Final pH	0.20	0.62
Moisture loss	2.42	7.52%
Dry matter loss	0.84	2.62%
Digestibility	0.78	2.44%

APPENDIX 2.

Second Batch Experiment

RESULTS AND STATISTICAL ANALYSES

SECOND BATCH EXPERIMENT RESULTS

Batch	1	2	3	4	5	6	7	8	9
Propionic acid treatment %	2.6	2.7	2.4	5.3	4.5	4.8	nil	nil	1.9
Initial moisture content %	39.5	38.2	32.7	32.5	36.9	34.9	28.3	26.2	24.4
Initial fresh weight kg	1435.2	1422.5	1416.1	1342.2	1305.0	1371.7	1181.1	1161.2	1232.0
Initial dry matter weight kg	868.3	879.1	953.0	906.0	823.5	893.0	846.8	850.0	931.4
Rate of baling kg per minute	163.4	160.4	175.2	80.5	95.4	100.2	-	-	221.0
Rate of acid application g/min.	4272	4272	4272	4272	4272	4800	-	-	4272
Initial W.S.C. % in dry matter	22.3	19.7	21.6	21.6	20.1	21.4	19.1	18.2	16.5
Final fresh weight kg	1131.0	1108.1	1172.3	1112.2	1085.6	1127.5	924.2	949.1	1073.4
Final dry matter %	72.83	75.21	74.89	75.65	74.93	73.34	83.49	84.19	81.08
Final dry matter weight kg	823.7	833.4	877.9	838.3	813.4	826.9	771.6	799.0	870.3
Dry matter loss %	5.14	5.20	7.88	7.47	1.24	7.40	8.88	5.94	6.56
Final W.S.C. % in dry matter	16.2	13.3	13.4	15.2	10.1	16.9	10.9	7.1	18.3
W.S.C. loss %	31.1	36.0	42.9	34.9	50.4	26.9	48.0	63.3	+3.6
Moisture loss %	45.79	49.45	36.43	37.92	43.48	37.21	54.35	50.68	32.51

SECOND BATCH EXPERIMENT RESULTS (cont.)

Batch	10	11	12	13	14	15	16	17	18
Propionic acid treatment %	2.3	2.1	3.1	3.3	3.2	5.0	4.9	5.0	nil
Initial moisture content %	27.8	25.2	24.8	23.1	25.7	24.8	24.0	24.0	21.3
Initial fresh weight kg	1113.6	1246.9	1127.2	1087.3	1121.7	1058.7	1085.9	1066.8	1085.4
Initial dry matter weight kg	804.0	932.7	847.7	836.1	833.4	796.1	825.3	810.8	854.2
Rate of baling kg per minute	184.1	206.1	139.7	130.5	134.6	96.0	98.4	96.7	-
Rate of acid application g/min	4272	4272	4272	4272	4272	4800	4800	4800	-
Initial W.S.C. % in dry matter	15.8	18.0	17.4	16.6	18.6	17.5	18.8	17.3	16.3
Final Fresh weight kg	1055.2	1083.2	1095.7	1026.5	1027.2	1022.7	1032.6	1041.3	1000.3
Final dry matter %	78.50	78.34	77.17	77.19	78.08	78.70	77.50	79.45	84.56
Final dry matter weight kg	828.3	848.6	845.6	792.4	802.0	804.9	800.3	827.3	845.9
Dry matter loss %	+3.02	9.02	0.25	5.23	3.77	+1.01	3.03	+2.04	1.00
Final W.S.C. % in dry matter	15.7	16.0	15.8	17.5	18.9	21.1	16.5	17.1	15.6
W.S.C. loss %	+2.4	19.1	9.4	0.1	2.2	+21.9	14.9	+0.9	5.2
Moisture loss %	26.72	25.33	10.52	6.77	21.89	17.03	10.85	16.42	33.20

SECOND BATCH EXPERIMENT CHANGES DURING STORAGE

Percentage dry matter

Batch	Initial	2 weeks	6 weeks	Final*
1	60.5	64.1	67.6	72.8
2	61.8	65.9	71.3	75.2
3	67.3	73.6	71.7	74.9
4	67.5	67.4	68.5	75.7
5	63.1	68.5	73.1	74.9
6	65.1	68.6	68.0	73.3
7	71.7	84.2	84.3	83.5
8	73.2	82.3	82.7	84.2
9	75.6	76.3	77.4	81.1
10	72.2	74.5	78.3	78.5
11	74.8	76.1	78.0	78.3
12	75.2	76.1	76.2	77.2
13	76.9	78.5	77.7	77.2
14	74.3	75.6	76.1	78.1
15	75.2	76.1	78.0	78.7
16	76.0	76.9	78.0	78.7
17	76.0	78.1	79.3	79.5
18	78.7	80.6	82.7	84.6

* 16 weeks

SECOND BATCH EXPERIMENT CHANGES DURING STORAGE

Water soluble carbohydrate

per cent in dry matter

Batch	Initial	2 weeks	6 weeks	Final*
1	22.3	21.6	17.3	16.2
2	19.7	19.0	13.4	13.3
3	21.6	20.6	18.9	13.4
4	21.6	21.9	16.9	15.2
5	20.1	21.1	17.6	10.1
6	21.4	20.4	18.1	16.9
7	19.1	15.2	15.2	10.9
8	18.2	15.5	17.1	7.1
9	16.5	17.1	19.0	18.3
10	15.8	21.0	16.3	15.7
11	18.0	15.7	17.6	16.0
12	17.4	16.5	17.9	15.8
13	16.6	16.1	16.6	17.5
14	18.6	17.6	18.5	18.9
15	17.5	16.7	18.2	21.1
16	18.8	20.9	17.3	16.5
17	17.3	20.1	17.4	17.1
18	16.3	18.5	19.2	15.6

* 16 weeks

TABLE 15

SECOND BATCH EXPERIMENT MICROBIOLOGICAL ANALYSES

Colony counts from the 2 week and 6 week samplings using the Air Displacement Sampler

	2 week sampling					6 week sampling				
	27°C		36°C		55°C	27°C		36°C		55°C
Sample	MSA	NA	MSA	NA	NA	MSA	NA	MSA	NA	NA
1a	15	15	15	0	2	48	2	39	1	0
1b	21	3	7	12	1	51	5	12	16	2
2a	26	17	6	4	500	45	9	7	4	5
2b	18	36	15	27	3	54	8	16	3	1
3a	11	16	10	5	4	38	2	3	0	1
3b	14	15	5	9	0	10	2	2	3	2

TABLE 15 ctd.

Sample	2 week sampling					6 week sampling				
	27°C		36°C		55°C	27°C		36°C		55°C
	MSA	NA	MSA	NA	NA	MSA	NA	MSA	NA	NA
4a	16	10	12	13	1	5	248	6	570	2
4b	4	11	8	8	3	2	415	3	195	5
5a	30	22	4	6	2	27	24	6	6	1
5b	10	9	3	6	4	62	6	9	4	5
6a	7	1	6	34	7	35	0	7	1	7
6b	18	24	3	33	1	6	4	1	0	3
7a	500	98	550	13	5	540	363	200	5	0
7b	500	161	512	0	2	260	122	220	4	3
8a	123	52	170	35	0	500	24	140	9	2
8b	172	108	56	7	0	600	8	430	8	1

TABLE 15 ctd.

Sample	2 week sampling					6 week sampling				
	27°C		36°C		55°C	27°C		36°C		55°C
	MSA	NA	MSA	NA	NA	MSA	NA	MSA	NA	NA
9a	53	15	11	4	2	25	5	11	0	3
9b	56	22	17	6	1	16	2	3	1	0
10a	11	23	15	16	0	128	31	27	1	3
10b	24	2	9	13	1	63	16	50	3	4
11a	31	13	20	22	0	71	20	36	1	8
11b	10	23	12	4	2	148	56	66	4	0
12a	43	8	9	15	1	16	6	10	2	2
12b	23	17	22	5	0	6	5	1	1	1
13a	52	2	11	7	0	0	1	4	5	13
13b	55	13	17	4	0	5	7	1	6	1
14a	43	7	9	7	0	3	2	0	3	1
14b	48	6	16	12	0	5	5	1	2	1

TABLE 15 ctd.

Sample	2 week sampling					6 week sampling				
	27°C		36°C		55°C	27°C		36°C		55°C
	MSA	NA	MSA	NA	NA	MSA	NA	MSA	NA	NA
15a	39	1	14	3	1	5	1	1	1	6
15b						3	7	2	3	0
16a	47	12	18	11	0	4	2	1	3	4
16b						6	1	2	4	2
17a	51	2	15	13	1	5	8	0	2	2
17b						5	10	6	3	0
18a	36	58	16	155	162	100	13	7	10	2
18b	43	53	12	141	235	45	26	10	1	1

SECOND BATCH EXPERIMENT

ANALYSIS OF VARIANCE

Percentage Water Soluble Carbohydrate Losses

Blocks	nil	2%	3%	5%	total	
A	48.0	19.1	2.2	+21.9	47.4	
B	63.3	+3.6	9.4	+0.9	68.2	
C	--	+2.4	0.1	14.9	12.6	
Total	111.3	13.1	11.7	+7.9	128.2	
Mean	55.7	4.4	3.9	+2.6		11.7

Correction for the mean 1494.1

Analysis of variance table

Source	df	Sum of squares	Mean Square	V. ratio
Blocks	2	283.3		
Treatments	3	4823.4	1607.8	9.0
Error	5	889.3	177.9	
Total	10	5996.0		

Standard error of nil treatment mean $= \sqrt{\frac{177.9}{2}} = 9.43$

Standard error of acid treatment means $= \sqrt{\frac{177.9}{3}} = 7.70$

Least significant difference between nil and other means (P = 0.05)

$= (9.43 + 7.70) \times 2.57$

$= 44.03$

The other results were analysed in the same way with the following values being found:-

	S.E. of nil	S.E. of acid	L.S.D.
Final moisture content	(no significant differences)		
Final W.S.C.	1.32	1.08	6.17
Dry matter loss	(no significant differences)		
Degree days above ambient	93.5	76.4	436.5

APPENDIX 3.

First Mini-bale Experiment

RESULTS AND STATISTICAL ANALYSES

TABLE 19 - FIRST MINI-BALE EXPERIMENT 1971

Results Initial Water Soluble Carbohydrate 13.74

ACID TREATMENT		NIL				1%			
Bale Number		3	7	16	23	6	11	14	21
Block Number		1	2	3	4	1	2	3	4
Initial bale weight	gm	1142	1154	1201	1172	1305	1210	1127	1115
Final bale weight	gm	499.5	504.2	533.0	544.4	602.6	518.1	519.4	518.9
Initial dry matter	%	50.4	50.4	50.4	50.4	51.0	51.0	51.0	51.0
Final dry matter	%	78.3	81.8	73.7	83.0	76.0	70.8	77.9	82.3
Dry matter loss	%	32.0	29.7	35.1	23.5	35.9	34.6	29.6	24.9
Moisture loss	%	80.85	83.97	76.47	84.08	78.93	71.93	79.21	83.19
Final W.S.C.	%	0.66	0.69	0.43	0.59	0.80	0.85	0.70	1.10
W.S.C. loss	%	96.7	96.5	98.0	96.7	96.3	96.0	96.4	94.0
Initial pH		6.5	6.5	6.5	6.5	5.5	5.5	5.5	5.5
Final pH		8.6	7.7	8.7	8.5	9.3	8.1	7.8	7.9
Maximum Temperature	^{o}C	32.7	30.4	30.5	32.4	42.5	37.8	34.8	35.1
Days to Max. Temp.		8	8	7	7	14	14	14	14

TABLE 19 ctd.

ACID TREATMENT		2%				4%			
Bale Number		2	9	18	22	5	10	15	24
Block Number		1	2	3	4	1	2	3	4
Initial bale weight	gm	1234	1270	1245	1252	1289	1189	1277	1254
Final bale weight	gm	605.5	637.0	615.6	635.8	1146	1180	1066	995.4
Initial dry matter	%	51.1	51.1	51.1	51.1	50.2	50.2	50.2	50.2
Final dry matter	%	70.4	75.6	77.3	81.0	56.4	55.1	58.1	56.6
Dry matter loss	%	32.4	25.8	25.2	19.5	0.1	+0.5	3.4	10.5
Moisture loss	%	70.30	74.74	77.05	80.27	22.15	17.45	29.78	30.82
Final W.S.C.	%	0.62	0.62	0.68	1.35	14.59	15.68	16.81	15.65
W.S.C. loss	%	97.0	98.3	96.3	92.1	+6.1	+14.7	+18.2	+2.0
Initial pH		5.0	5.0	5.0	5.0	4.6	4.6	4.6	4.6
Final pH		8.9	8.5	9.0	8.0	4.9	4.9	4.9	4.9
Maximum Temperature	°C	31.5	29.7	30.6	34.9	21.9	17.8	18.1	19.7
Days to Max. Temp.		25	27	27	23	15	20	20	20

TABLE 19 ctd.

ACID TREATMENT		6%				8%			
Bale Number		1	12	13	19	4	8	17	20
Block Number		1	2	3	4	1	2	3	4
Initial bale weight	gm	1272	1282	1247	1252	1289	1253	1273	1181
Final bale weight	gm	1047	1697	1029	9535	1143	1104	1087	1009
Initial dry matter	%	51.3	51.3	51.3	51.3	50.9	50.9	50.9	50.9
Final dry matter	%	57.2	56.6	55.2	58.6	54.0	57.1	55.5	57.4
Dry matter loss	%	6.2	5.6	11.2	13.0	5.9	1.2	6.9	3.7
Moisture loss	%	26.07	23.75	24.08	35.25	16.90	23.05	22.62	25.91
Final W.S.C.	%	12.80	14.94	13.51	13.06	14.42	15.36	12.95	15.05
W.S.C. loss	%	12.6	+2.7	12.7	17.3	1.2	+10.5	12.2	+5.5
Initial pH		4.4	4.4	4.4	4.4	4.3	4.3	4.3	4.3
Final pH		4.8	4.6	4.7	4.8	4.6	4.9	4.7	4.7
Maximum Temperature	°C	17.8	18.0	17.9	19.0	18.5	18.6	18.9	20.5
Days to Max. Temp.		20	20	20	20	20	20	20	20

FIRST MINI-BALE EXPERIMENT

ANALYSIS OF VARIANCE

Dry Matter Losses

Blocks	nil	1%	2%	4%	6%	8%	Total
1	32.0	35.9	32.4	0.1	6.2	5.9	112.5
2	29.7	34.6	25.8	+0.5	5.6	1.2	96.4
3	35.1	29.6	25.2	3.4	11.2	6.9	111.4
4	23.5	24.9	19.5	10.5	13.0	3.7	95.1
Total	120.3	125.0	102.9	13.5	36.0	17.7	415.4
Mean	30.1	31.3	25.7	3.4	9.0	4.4	17.3

Correction for the mean 7189.88

Analysis of Variance Table

Source	df	Sum of sqs.	Mean sq.	Variance ratio
Block	3	43.98	14.66	
Treatment	5	3429.38	685.88	31.77
Error	15	323.80	21.59	
Total	23	3797.16		

Standard error of means $= \sqrt{\frac{21.59}{4}} = 2.32$

Standard error of difference between means $= \sqrt{\frac{21.59}{2}} = 3.29$

Least significant difference between means (P = 0.01)

$= 3.29 \times 2.95 = 9.71\ \%$

Analyses of variance for the other results of the experiment were carried out in the same way and the following values were obtained:-

	S.E. between means	L.S.D. (P = 0.01)
Percentage moisture loss	2.47	7.29
Percentage W.S.C. loss	4.37	12.90
Degree days above ambient	11.83	34.92

APPENDIX 4.

Second Mini-bale Experiment

RESULTS AND STATISTICAL ANALYSES

SECOND MINI-BALE EXPERIMENT

Moisture Level 76%

Initial Water Soluble Carbohydrate 28.25%

Acid Treatment		Nil				3.6%			
Bale Number		1	8	13	19	6	12	18	24
Block Number		1	2	3	4	1	2	3	4
Initial Bale weight	gm.	2980	2990	2817	2715	2799	2721	2790	2862
Final Bale weight	gm.	1488	849	1363	1134	2481	1970	2493	2316
Initial dry matter	%	22.18	22.18	22.18	22.18	24.70	24.70	24.70	24.70
Final dry matter	%	16.59	29.73	18.15	21.31	25.75	30.11	26.01	28.58
Dry matter loss	%	62.65	61.94	60.41	59.87	7.59	11.73	5.91	6.36
Moisture loss	%	46.48	74.36	49.11	57.77	12.60	32.80	12.20	23.25
Final W.S.C. content	%	0.58	1.06	1.04	0.68	28.15	22.11	21.46	28.90
W.S.C. loss	%	99.24	98.58	98.54	99.03	7.91	30.91	28.53	4.21
Initial pH		5.6	5.6	5.6	5.6	4.4	4.4	4.4	4.4
Final pH		8.0	8.26	-	8.17	4.18	4.2	4.0	4.2
Degree days above ambient		326.4	236.7	282.5	263.8	35.7	25.7	28.0	2.2

SECOND MINI-BALE EXPERIMENT

Moisture Level 76%

Initial Water Soluble Carbohydrate 28.25%

Acid Treatment	4.7%				5.6%			
Bale Number	5	11	17	23	4	10	16	22
Block Number	1	2	3	4	1	2	3	4
Initial Bale weight gm.	2769	2754.5	2754	2748	2727	2711	2722	2681
Final Bale weight gm.	2233	2280	2336	2131	2243	2243	2360	1856
Initial dry matter %	23.34	23.34	23.34	23.34	25.14	25.14	25.14	25.14
Final dry matter %	27.18	27.64	26.60	29.55	26.38	27.21	25.55	32.27
Dry matter loss %	6.09	1.95	3.33	1.82	13.70	10.44	11.88	11.14
Moisture loss %	23.39	21.87	18.79	28.73	19.11	19.55	13.77	37.37
Final W.S.C. content %	28.15	12.85	27.53	17.87	24.54	20.25	23.26	19.92
W.S.C. loss %	18.43	55.41	5.78	37.90	27.81	35.81	27.44	37.34
Initial pH	4.1	4.1	4.1	4.1	4.1	4.1	4.1	4.1
Final pH	4.0	4.1	4.0	4.2	4.0	4.0	4.0	4.1
Degree days above ambient	46.3	14.0	12.8	12.7	1.5	2.4	55.0	-9.1

SECOND MINI-BALE EXPERIMENT

Moisture Level 76%

Initial Water Soluble Carbohydrate 28.25%

Acid Treatment	6.7%				7.6%			
Bale Number	3	7	15	21	2	9	14	20
Block Number	1	2	3	4	1	2	3	4
Initial Bale weight gm.	2689	2705	2731.5	2748	2729	2617	2631	2677
Final Bale weight gm.	2353	2271	2491	2093	2357	2016	2364	1918
Initial dry matter %	23.79	23.79	23.79	23.79	26.60	26.60	26.60	26.60
Final dry matter %	25.70	25.85	24.92	29.38	26.48	29.25	25.44	30.96
Dry matter loss %	5.47	8.76	4.47	5.93	14.02	15.29	14.06	16.62
Moisture loss %	14.69	18.31	10.16	29.42	13.49	25.75	8.73	32.61
Final W.S.C. content %	25.95	11.22	22.40	20.44	15.88	15.74	21.66	29.21
W.S.C. loss %	13.17	63.77	24.25	31.98	51.68	52.88	34.11	13.78
Initial pH	4.0	4.0	4.0	4.0	4.0	4.0	4.0	4.0
Final pH	3.9	3.9	3.8	4.1	4.0	3.9	4.0	4.0
Degree days above ambient	37.8	35.6	10.4	-4.4	28.2	2.4	74.0	-0.6

SECOND MINI-BALE EXPERIMENT

Moisture Level 62%

Initial Water Soluble Carbohydrate 29.80%

Acid Treatment	Nil				3.1%			
Bale Number	25	31	37	43	30	36	42	48
Block Number	1	2	3	4	1	2	3	4
Initial Bale weight gm.	1890	1732	1731	1979	1986	2027	2079	2070
Final Bale weight gm.	879	558	684	671	1747	1585	1852	1715
Initial dry matter %	38.58	38.58	38.58	38.58	41.84	41.84	41.84	41.84
Final dry matter %	35.86	54.50	39.45	51.60	43.74	44.16	44.88	46.85
Dry matter loss %	56.77	54.49	59.59	54.65	8.03	17.45	4.45	7.21
Moisture loss %	51.43	76.13	61.04	73.28	14.91	24.92	15.57	24.29
Final W.S.C. content %	1.51	0.78	0.37	0.45	30.40	13.23	33.21	26.11
W.S.C. loss %	97.81	98.81	99.50	99.31	6.19	63.35	+6.45	18.68
Initial pH	5.6	5.6	5.6	5.6	4.7	4.7	4.7	4.7
Final pH	8.7	8.7	8.7	8.7	4.4	5.3	4.5	4.5
Degree days above ambient	382.8	336.5	373.4	404.6	5.9	34.9	44.0	34.7

SECOND MINI-BALE EXPERIMENT

Moisture Level 62%

Initial Water Soluble Carbohydrate 29.80%

Acid Treatment	3.6%				4.2%			
Bale Number	29	35	41	47	28	34	40	46
Block Number	1	2	3	4	1	2	3	4
Initial Bale weight gm.	2028.5	2071.5	2035	2051.5	1983	2054	2032.5	2059
Final Bale weight gm.	1819	1733	1792	1616	1636	1680	1810	1754
Initial dry matter %	41.01	41.01	41.01	41.01	40.03	40.03	40.03	40.03
Final dry matter %	41.81	44.88	44.10	44.49	46.18	46.59	43.84	43.50
Dry matter loss %	8.58	8.45	5.31	14.54	4.82	4.80	2.47	7.43
Moisture loss %	11.54	21.83	16.55	25.88	25.96	27.16	16.60	19.74
Final W.S.C. content %	30.34	27.35	31.47	29.55	30.69	29.16	29.41	30.56
W.S.C. loss %	6.93	15.98	Nil	15.26	1.98	6.85	3.75	5.04
Initial pH	4.6	4.6	4.6	4.6	4.3	4.3	4.3	4.3
Final pH	4.4	4.4	4.5	4.4	4.4	4.3	4.3	4.4
Degree days above ambient	22.6	2.0	1.0	-11.5	26.2	-6.0	-9.1	-13.5

SECOND MINI-BALE EXPERIMENT

Moisture Level 62%

Initial Water Soluble Carbohydrate 29.80%

Acid Treatment	4.6%				5.0%			
Bale Number	27	33	39	45	26	32	38	44
Block Number	1	2	3	4	1	2	3	4
Initial Bale weight gm.	2088	2034	1969	2032	2004	2052	2062	2117
Final Bale weight gm.	1762	1570	1676	1676	1757	1646	1743.5	1770.5
Initial dry matter %	43.37	43.37	43.37	43.37	41.66	41.66	41.66	41.66
Final dry matter %	44.28	50.64	45.49	45.54	40.35	47.43	44.22	43.80
Dry matter loss %	13.85	9.87	10.71	13.40	15.09	8.68	10.24	12.07
Moisture loss %	16.82	32.72	18.06	20.68	10.36	27.72	19.16	19.44
Final W.S.C. content %	21.53	27.31	24.09	28.87	27.26	28.03	30.39	34.58
W.S.C. loss %	37.76	17.41	27.83	16.10	22.33	14.10	8.47	+2.04
Initial pH	4.5	4.5	4.5	4.5	4.3	4.3	4.3	4.3
Final pH	4.4	4.4	4.3	4.3	4.3	4.3	4.2	4.3
Degree days above ambient	14.8	-26.6	9.3	-9.6	-3.7	5.2	-5.0	-10.4

SECOND MINI-BALE EXPERIMENT

Moisture Level 52%

Initial Water Soluble Carbohydrate 24.03%

Acid Treatment	Nil				2.2%			
Bale Number	49	55	61	67	54	60	66	72
Block Number	1	2	3	4	1	2	3	4
Initial Bale weight gm.	1487	1488	1488	1491	1419	1458	1415	1444
Final Bale weight gm.	537	549	617	669	1256	1169	1205	1294
Initial dry matter %	47.29	47.29	47.29	47.29	52.84	52.84	52.84	52.84
Final dry matter %	83.71	82.31	82.34	82.00	57.92	61.65	60.37	57.33
Dry matter loss %	31.79	35.78	27.80	22.18	2.99	6.49	2.71	2.77
Moisture loss %	88.84	87.62	86.11	84.68	21.03	34.80	28.44	18.92
Final W.S.C. content %	Nil	Nil	7.65	Nil	27.77	19.23	27.70	27.32
W.S.C. loss %	100	100	77.02	100	+12.11	25.17	+12.16	+10.55
Initial pH	6.50	6.50	6.50	6.50	5.20	5.20	5.20	5.20
Final pH	-	8.0	-	-	4.60	4.67	4.59	4.72
Degree days above ambient	357.7	349.3	309.6	391.3	0.1	-10.2	20.3	2.4

SECOND MINI-BALE EXPERIMENT

Moisture Level 52%

Initial Water Soluble Carbohydrate 24.03%

Acid Treatment	2.75%				3.21%			
Bale Number	53	59	65	71	52	58	64	70
Block Number	1	2	3	4	1	2	3	4
Initial Bale weight gm.	1444	1522	1508	1521	1476	1455	1475	1492
Final Bale weight gm.	1230	1333	1380	1323	1251	1233	1284	1343
Initial dry matter %	53.63	53.63	53.63	53.63	53.25	53.25	53.25	53.25
Final dry matter %	61.26	59.93	56.81	59.30	61.58	62.79	61.10	58.22
Dry matter loss %	2.66	2.10	3.06	3.82	1.99	0.07	0.11	1.59
Moisture loss %	28.84	24.32	14.76	23.65	30.35	32.55	27.57	19.56
Final W.S.C. content %	25.91	25.78	21.88	26.77	26.81	26.12	25.63	27.27
W.S.C. loss %	+4.95	+5.04	11.73	+7.14	+9.35	+8.62	+6.54	+11.68
Initial pH	4.95	4.95	4.95	4.95	4.79	4.79	4.79	4.79
Final pH	4.63	4.65	4.61	4.60	4.57	4.58	4.60	4.63
Degree days above ambient	0.1	20.3	28.4	28.2	13.2	22.1	52.6	32.4

SECOND MINI-BALE EXPERIMENT

Moisture Level 52%

Initial Water Soluble Carbohydrate 24.03%

Acid Treatment	3.75%				4.20%			
Bale Number	51	57	63	69	50	56	62	68
Block Number	1	2	3	4	1	2	3	4
Initial Bale weight gm.	1592	1495	1456	1500	1500	1486	1449	1495
Final Bale weight gm.	1387	1206	1254	1388	1287	1210	1290	1341
Initial dry matter %	53.60	53.60	53.60	53.60	51.94	51.94	51.94	51.94
Final dry matter %	59.81	60.66	59.49	54.99	59.95	63.57	59.87	56.88
Dry matter loss %	2.78	8.74	4.41	5.07	0.93	0.34	+2.62	1.73
Moisture loss %	24.54	31.61	24.81	10.24	28.49	38.28	25.66	19.52
Final W.S.C. content %	27.60	20.59	21.70	20.88	20.88	27.96	26.12	26.42
W.S.C. loss %	+11.67	21.80	13.68	17.51	13.91	+15.96	+11.71	+8.05
Initial pH	4.80	4.80	4.80	4.80	4.59	4.59	4.59	4.59
Final pH	4.57	-	4.57	4.51	4.50	-	4.63	4.58
Degree days above ambient	11.3	2.6	4.0	11.7	27.2	10.9	144.0	16.2

SECOND MINI-BALE EXPERIMENT

Moisture Level 41%

Initial Water Soluble Carbohydrate 24.25%

Acid Treatment	Nil				1.0%			
Bale Number	73	79	85	91	78	84	90	96
Block Number	1	2	3	4	1	2	3	4
Initial Bale weight gm.	1226	1238	1239	1250	1238	1238	1222	1254
Final Bale weight gm.	647	615	629	639	835	741	732	862
Initial dry matter %	58.98	58.98	58.98	58.98	58.75	58.75	58.75	58.75
Final dry matter %	81.12	79.30	80.62	81.44	78.39	73.75	78.53	72.61
Dry matter loss %	27.42	33.2	30.66	29.41	10.00	24.90	19.93	15.58
Moisture loss %	75.71	74.93	76.02	76.87	64.67	61.91	68.82	54.36
Final W.S.C. content %	3.75	5.66	4.99	2.32	6.16	7.15	3.21	Nil
W.S.C. loss %	88.78	84.41	85.74	93.25	77.14	77.85	89.40	100.0
Initial pH	6.00	6.00	6.00	6.00	5.60	5.60	5.60	5.60
Final pH	8.11	8.43	7.99	8.31	6.94	8.10	7.99	6.89
Degree days above ambient	239.5	222.9	211.4	207.0	90.3	124.9	108.5	42.4

SECOND MINI-BALE EXPERIMENT

Moisture Level 41%

Initial Water Soluble Carbohydrate 24.25%

Acid Treatment	1.5%				2.0%			
Bale Number	77	83	89	95	76	82	88	94
Block Number	1	2	3	4	1	2	3	4
Initial Bale weight gm.	1236	1261	1240	1254	1240	1237	1246	1221
Final Bale weight gm.	1047	1121	1102	1074	1073	1078	1061	1085
Initial dry matter %	59.50	59.50	59.50	59.50	58.45	58.45	58.45	58.45
Final dry matter %	70.71	66.94	66.15	68.48	68.41	66.81	70.23	66.43
Dry matter loss %	+0.67	+0.01	1.03	1.42	+1.27	0.39	+2.31	+0.94
Moisture loss %	38.74	27.43	25.72	33.34	34.21	30.39	39.19	28.20
Final W.S.C. content %	19.98	19.37	11.15	15.70	16.95	17.51	23.02	14.28
W.S.C. loss %	17.06	20.11	54.49	36.18	29.21	28.07	2.88	40.56
Initial pH	5.30	5.30	5.30	5.30	5.19	5.19	5.19	5.19
Final pH	5.11	5.08	5.00	5.00	5.01	4.81	5.27	4.92
Degree days above ambient	30.9	22.9	39.5	45.1	63.7	7.6	45.4	26.7

SECOND MINI-BALE EXPERIMENT

Moisture Level 41%

Initial Water Soluble Carbohydrate 24.25%

Acid Treatment	2.5%				3.0%			
Bale Number	75	81	87	93	74	80	86	92
Block Number	1	2	3	4	1	2	3	4
Initial Bale weight gm.	1236	1250	1243	1229	1244	1243	1246	1250
Final Bale weight gm.	1078	1072	1077	1072	1027	1067	1081	1129
Initial dry matter %	57.90	57.90	57.90	57.90	59.00	59.00	59.00	59.00
Final dry matter %	68.62	68.64	67.82	67.75	70.40	68.58	67.53	64.93
Dry matter loss %	+3.37	+1.67	+1.48	+2.06	1.50	0.27	0.69	0.60
Moisture loss %	34.99	36.12	33.77	33.18	40.40	34.22	31.29	22.74
Final W.S.C. content %	18.28	20.19	22.07	19.14	15.27	11.84	19.04	22.02
W.S.C. loss %	22.08	15.35	7.64	19.44	37.98	51.31	22.03	9.74
Initial pH	4.99	4.99	4.99	4.99	4.89	4.89	4.89	4.89
Final pH	4.97	-	4.93	4.78	4.75	4.89	4.73	4.62
Degree days above ambient	34.9	19.1	50.1	8.6	30.9	33.1	9.5	62.9

SECOND MINI-BALE EXPERIMENT

Moisture Level 31%

Initial Water Soluble Carbohydrate 24.90%

Acid Treatment	Nil				0.5%			
Bale Number	97	103	109	115	101	107	113	119
Block Number	1	2	3	4	1	2	3	4
Initial Bale weight gm.	1070	1091	1097	1009	1092	1087	1111	1108
Final Bale weight gm.	604	715	728	715	664	763	845	785
Initial dry matter %	66.96	66.96	66.96	66.96	69.80	69.80	69.80	69.80
Final dry matter %	81.12	80.56	81.56	81.57	81.99	82.94	80.11	82.01
Dry matter loss %	31.61	21.15	19.17	13.68	28.57	16.59	12.71	16.76
Moisture loss %	67.74	61.44	62.96	60.47	63.74	60.35	49.90	57.80
Final W.S.C. content %	2.20	2.22	2.70	2.83	4.35	3.42	14.29	4.10
W.S.C. loss %	93.95	92.97	91.24	90.19	87.52	88.54	49.91	86.29
Initial pH	6.10	6.10	6.10	6.10	5.90	5.90	5.90	5.90
Final pH	7.60	7.61	5.27	6.90	6.94	6.84	5.87	6.99
Degree days above ambient	126.1	139.7	102.2	96.9	116.4	80.5	50.2	68.5

SECOND MINI-BALE EXPERIMENT

Moisture Level 31%

Initial Water Soluble Carbohydrate 24.90%

Acid Treatment	1.0%				1.5%			
Bale Number	100	106	112	118	99	105	111	117
Block Number	1	2	3	4	1	2	3	4
Initial Bale weight gm.	1105	1091	1077	1108	1115	1084	1090	1100
Final Bale weight gm.	965	930	933	962	959	967	985	963
Initial dry matter %	69.60	69.60	69.60	69.60	66.75	66.75	66.75	66.75
Final dry matter %	76.02	75.27	74.74	75.41	76.2	75.15	74.85	75.67
Dry matter loss %	4.62	7.81	7.27	5.93	1.80	+0.43	+1.33	0.76
Moisture loss %	31.40	30.66	28.02	29.77	38.44	33.33	31.65	35.94
Final W.S.C. content %	20.91	16.07	15.77	14.89	17.36	7.98	19.12	20.30
W.S.C. loss %	19.90	40.50	41.27	43.75	31.54	67.82	22.19	19.10
Initial pH	5.75	5.75	5.75	5.75	5.51	5.51	5.51	5.51
Final pH	5.57	5.70	5.75	5.83	5.63	5.58	5.60	5.83
Degree days above ambient	26.6	11.4	33.1	47.3	12.0	19.6	18.3	24.9

SECOND MINI-BALE EXPERIMENT

Moisture Level 31%

Initial Water Soluble Carbohydrate 24.90%

Acid Treatment		2.0%			
Bale Number		98	104	110	116
Block Number		1	2	3	4
Initial Bale weight	gm.	1102	1085	1158	1093
Final Bale weight	gm.	980	973	1053	984
Initial dry matter	%	69.02	69.02	69.02	69.02
Final dry matter	%	76.19	74.32	73.80	74.72
Dry matter loss	%	1.83	3.44	2.77	2.54
Moisture loss	%	28.78	25.66	23.10	26.54
Final W.S.C. content	%	20.90	23.58	20.66	12.62
W.S.C. loss	%	17.60	8.56	19.33	50.61
Initial pH		5.31	5.31	5.31	5.31
Final pH		7.16	5.08	5.36	5.22
Degree days above ambient		6.7	-14.7	10.3	23.9

SECOND MINI-BALE EXPERIMENT

ANALYSIS OF VARIANCE

Dry matter losses at 30 per cent moisture level

Blocks	nil	0.5%	1.0%	1.5%	2.0%	Total
1	31.61	28.57	4.62	1.80	1.83	68.43
2	21.15	16.59	7.81	+0.43	3.44	48.56
3	19.17	12.71	7.27	+1.33	2.77	40.59
4	13.68	16.76	5.93	0.76	2.54	39.67
Total	85.61	74.63	25.63	0.80	10.58	197.25
Mean	24.40	18.66	6.41	0.20	2.65	9.86

Correction for the mean 1945.38

Analysis of variance table

Source	df	Sum of Sqs.	Mean Sq.	Var. Ratio
Block	3	107.02	35.67	
Treatment	4	1471.67	367.92	20.39
Error	12	216.43	18.04	
Total	19	1795.12		

Standard Error of differences between means $= \sqrt{\frac{18.04}{2}}$

$= 3.00$

Least significant difference (P = 0.01) $= 3.00 \times 3.05$

$= 9.16$ per cent

Analyses of variance were carried out on the rest of the results in a similar way and the values for the least significant differences are given along with the results in Tables 21 and 22.

APPENDIX 5.

First Distribution Experiment

RESULTS AND STATISTICAL ANALYSES

FIRST DISTRIBUTION EXPERIMENT

SWATH MEASUREMENTS (per metre)

Weights	Heights	Widths
2119 gm	30 cm	63 cm
1569	33	65
1519	34	58
2494	24	70
1321	27	53
1944	24	63
1719	23	67
1594	24	50
2369	33	95
2419	37	56
1819	32	65
1394	22	70
2369	27	100
2544	32	85
1396	31	77
1844	35	80
2969	35	108
2469	34	74
1919	38	77
2294	22	71
1544	27	61
2094	38	74
2194	21	82
2294	41	97
3671	34	122
908	23	51
1554	33	82
2444	23	50
1994	23	60
1271	40	71
2069	43	58
1769	35	72
2469	35	104
2719	28	59
3144	48	60
1496	34	63
2044	34	78
2544	41	77
2819	47	69
3469	45	80
1944	37	85
2644	28	91
2369	54	73
833	21	53
3433	29	108
2644	38	100

46 metres

Weights	Heights	Widths
3119 gm	45 cm	97 cm
2819	33	57
1071	34	73
1544	24	82
2169	48	93
2369	31	86
2019	36	93
1869	10	64
1421	31	90
2044	31	79
1421	30	77
2394	32	80
1844	25	85
1769	36	78
2019	46	68
2119	35	55
1769	41	70
2344	30	84
2044	34	60
3183	45	62
1158	31	64
1158	37	80
2594	33	70
1619	36	70
1471	33	92
2019	39	54
1869	39	84
2144	24	68
1246	35	74
2019	26	80
1694	31	82
1719	38	108
2044	33	91
1944	35	82
2544	49	100
2641	39	68
2119	35	98
1819	37	94
2444	21	93
4083	53	100
2869	37	100
308	37	72
2069	35	78
3503	48	100
1003	28	84
1769	43	88

92 metres

SWATH MEASUREMENTS (cont.)

Weights	Heights	Widths
3503 gm	51 cm	61 cm
2269	47	72
3363	0	0
3119	40	130
2819	40	127
4333	32	89
3758	36	138
2794	27	129
2419	32	78
2119	28	76
2044	39	67
1108	34	58
2644	39	87
2919	24	68
1506	22	54
1496	31	66
3508	31	62
2369	34	72
2544	20	30
983	44	63
2869	23	50
1283	33	49
1719	32	64
1969	31	68
2919	49	89
1133	33	65
1669	46	92
2869	16	43
2644	33	88
2844	39	72
2844	33	83
633	33	82
1594	24	65
3519	53	92
3494	35	63
3144	56	94
1869	36	73
1869	32	87
2719	43	97
2469	41	81
2214	48	81
2394	32	51
2569	46	94
2169	43	62
1919	25	64
1844	58	68
2069	26	43
1156	47	76
2519	57	89
2594	47	80

142 metres

Weights

3663	
2519	
1994	
1844	
1433	
2669	
2169	
2344	
2619	
2169	
1919	
3656	
1944	

155 metres

Z	
833292075.0000 B0	sum x^2
79581176.8775 D0	
718.8608 A0	S.D.
2205.1419 C0	Mean
32.5992 B0	Co Variance
57.7407 A0	S.E.

Heights	Widths
41	81
38	61
46	77
24	32
31	57
39	84
41	97
36	52
30	73
41	78
39	93
44	95
23	65

Z	Z
199422.0000 B0	956757.0000 B0
12683.9355 D0	56758.8000 D0
9.0754 A0	19.1980 A0
34.7096 C0	76.2000 C0
26.1466 B0	25.1942 B0
0.7289 A0	1.5420 A0

FIRST DISTRIBUTION EXPERIMENT RESULTS FOR UNTREATED BALES

		Parts									
		1	2	3	4	5	6	7	8	9	Mean
Bale	Section	Percentage moisture content									
3	B	34.79	37.14	37.01	38.96	38.84	39.35	35.10	39.12	35.87	35.90
3	D	36.07	35.46	35.75	34.98	39.50	36.37	35.99	38.18	35.75	36.45
3	F	39.54	45.29	51.17	42.52	43.70	47.27	39.56	45.06	38.78	43.71
3	H	38.90	38.12	40.51	39.83	36.69	39.01	38.78	42.02	39.53	39.26
2	D	38.67	32.35	31.30	30.84	31.88	32.61	32.11	32.21	33.87	32.87
2	H	38.50	38.45	49.23	38.99	40.44	38.69	35.64	29.17	38.53	38.63

	Sections								
	A	B	C	D	E	F	G	H	Mean
Bale	Percentage Moisture content								
2	33.46	30.94	36.08	32.87	37.01	29.29	36.08	38.63	34.30
3	40.99	37.13	38.16	36.45	33.70	43.71	35.90	39.26	38.16

FIRST DISTRIBUTION EXPERIMENT RESULTS FOR BALE 5

	Parts 1	2	3	4	5	6	7	8	9	Mean
Sections	Percentage moisture content									
B	37.67	38.48	39.41	39.04	34.25	40.94	39.35	37.74	46.25	39.24
D	34.46	36.06	35.82	34.43	37.53	34.69	36.30	32.48	30.85	34.69
F	38.22	36.32	38.02	37.50	37.26	35.00	36.37	35.74	35.72	36.68
H	32.03	34.81	34.44	38.03	35.80	36.52	35.26	37.88	34.35	35.42
	pH values									
B	7.12	7.34	7.61	6.81	7.08	7.32	7.79	7.74	7.45	
D	4.71	4.89	4.70	4.72	5.37	4.75	4.80	5.59	4.90	
F	4.90	5.93	6.79	4.76	5.65	5.44	4.77	5.36	5.30	
H	4.65	6.01	5.78	4.75	5.75	5.26	4.87	5.13	5.00	
	Propionic acid level									
B	<0.50	<0.50	0.00	<0.50	<0.50	<0.50	0.00	0.00	0.00	<0.50
D	2.71	2.17	2.75	2.70	1.31	2.55	2.40	1.10	2.15	2.00
F	2.15	0.87	0.50	2.55	1.05	1.21	2.50	1.35	1.40	1.22
H	3.00	0.80	0.95	2.55	0.98	1.45	2.25	1.65	1.90	1.45

Sections A	B	C	D	E	F	G	H	Mean
Percentage moisture content								
38.92	39.24	35.97	34.69	37.55	36.68	36.25	35.42	36.84
pH values								
7.58	7.36	5.15	4.94	5.20	5.43	5.37	5.24	-
Propionic acid levels								
0.00*	<0.05*	1.60	2.00	1.55	1.22	1.31	1.45	1.52

* Not included in the mean

FIRST DISTRIBUTION EXPERIMENT RESULTS FOR BALE 10

	Parts									
	1	2	3	4	5	6	7	8	9	Mean
Sections	Percentage moisture content									
B	34.48	36.77	43.24	36.55	37.61	47.33	35.23	36.46	52.77	40.05
D	35.18	33.46	32.86	34.02	32.44	29.22	32.44	33.92	33.97	33.06
F	36.12	34.22	33.56	32.36	31.69	40.03	34.37	33.82	35.35	34.61
H	35.27	37.66	34.69	37.42	31.76	30.97	37.00	34.30	31.06	34.46
	pH value									
B	4.82	5.76	5.26	4.69	5.22	4.95	4.84	5.12	5.42	
D	4.62	5.14	5.51	4.87	5.66	5.50	4.75	5.07	5.14	
F	4.50	4.81	4.80	4.68	5.00	5.00	4.80	4.80	4.71	
H	4.50	4.92	4.78	4.70	5.29	4.96	4.88	4.93	4.81	
	Propionic acid level									
B	2.35	0.97	1.40	2.75	1.53	2.00	2.25	1.63	1.23	1.66
D	3.25	1.60	1.13	2.20	1.05	1.15	2.55	1.80	1.60	1.66
F	3.90	2.37	2.40	2.75	1.90	1.90	2.40	2.40	2.75	2.40
H	3.90	2.12	2.45	2.75	1.40	2.00	2.15	2.10	2.40	2.25

Sections									
A	B	C	D	E	F	G	H	Mean
Percentage moisture content									
31.87	40.05	35.00	33.06	33.33	34.61	34.04	34.46	34.68
pH values									
4.85	5.12	4.90	5.14	4.81	4.79	4.82	4.86	-
Propionic acid level									
2.25	1.68	2.15	1.60	2.40	2.40	2.40	2.25	2.14

APPENIDX 6.

Second Distribution Experiment

RESULTS AND STATISTICAL ANALYSES

SECOND DISTRIBUTION EXPERIMENT

SWATHS A & B — BALES 1, 2, 3 and 4.

Bale 1	Sections A	B	C	D	E	F	G	H	Mean
	Percentage moisture content								
	23.88	24.65	25.32	23.26	22.80	22.45	23.15	20.33	23.23
	Percentage propionic acid								
	3.89	4.60	5.68	4.29	3.15	4.26	3.96	3.90	4.22
Bale 2	Sections A	B	C	D	E	F	G	H	Mean
	Percentage moisture content								
	24.08	22.96	25.37	24.22	24.86	24.02	24.96	21.85	24.04
	Percentage propionic acid								
	3.59	3.17	2.98	4.42	2.61	2.94	2.63	2.76	3.14
Bale 3	Sections A	B	C	D	E	F	G	H	Mean
	Percentage moisture content								
	23.38	25.09	23.48	24.40	23.50	23.60	22.44	22.61	23.56
	Percentage propionic acid								
	4.11	3.96	1.97	4.28	3.11	1.82	2.80	6.59	3.58
Bale 4	Sections A	B	C	D	E	F	G	H	Mean
	Percentage moisture content								
	24.65	24.06	25.10	25.27	24.76	24.20	24.70	24.39	24.70
	Percentage propionic acid								
	2.56	3.30	7.92	4.77	3.75	2.70	2.12	3.22	3.79

SECOND DISTRIBUTION EXPERIMENT

SWATHS C & D BALES 5, 6, 7 and 8.

Bale 5	Sections A	B	C	D	E	F	G	H	Mean
	Percentage moisture content								
	28.80	28.36	28.90	25.79	29.13	28.65	30.59	27.03	28.41
	Percentage propionic acid								
	2.01	1.78	3.26	2.24	1.76	2.40	3.04	2.50	2.37
Bale 6	Sections A	B	C	D	E	F	G	H	Mean
	Percentage moisture content								
	29.47	29.79	28.58	30.80	34.65	31.33	30.01	30.00	30.58
	Percentage propionic acid								
	2.53	-	2.42	2.66	2.80	2.12	2.34	3.00	2.55
Bale 7	Sections A	B	C	D	E	F	G	H	Mean
	Percent moisture content								
	27.27	27.58	25.42	28.99	28.58	27.62	25.56	24.11	26.89
	Percent propionic acid								
	2.68	5.41	3.02	1.62	2.43	2.50	3.02	-	2.95
Bale 8	Sections A	B	C	D	E	F	G	H	Mean
	Percent moisture content								
	25.94	27.24	30.26	29.69	28.29	30.21	30.05	26.29	28.62
	Percentage propionic acid								
	2.89	1.62	3.74	2.96	3.05	0.78	4.02	3.43	2.81

SECOND DISTRIBUTION EXPERIMENT

SWATH A — BALES 1 and 2

Bale 1	Parts 1	2	3	4	5	6	7	8	9	mean
Sections	Percentage moisture content									
A	22.17	25.85	21.79	22.93	23.70	25.51	24.48	23.04	25.43	23.88
C	23.98	25.44	27.00	23.86	23.38	28.74	24.53	24.86	26.13	25.32
E	22.91	22.77	23.67	22.54	21.02	25.40	17.24	23.82	25.86	22.80
G	22.93	26.10	20.69	23.14	23.49	23.06	22.79	23.56	22.56	23.15
	Percentage propionic acid									
A	4.40	7.06	0.54	4.10	3.93	2.26	6.35	5.16	1.20	3.89
C	7.86	6.84	6.29	6.52	2.44	4.45	5.78	4.73	6.24	5.68
E	1.86	5.19	1.34	2.28	2.70	2.34	2.46	5.65	4.57	3.15
G	2.66	8.18	2.90	4.36	2.86	2.22	4.67	3.88	3.89	3.96

Bale 2	Parts 1	2	3	4	5	6	7	8	9	mean
Sections	Percentage moisture content									
A	24.60	24.94	26.13	22.33	25.10	23.88	21.88	23.97	23.90	24.08
c	22.08	30.28	26.31	24.31	25.66	24.58	27.13	24.15	23.80	25.37
E	24.11	28.12	24.30	24.30	24.18	25.12	25.39	24.71	23.49	24.86
G	23.82	26.81	22.85	24.30	25.89	25.38	24.84	26.84	23.89	24.96
	Percentage propionic acid									
A	4.33	6.06	1.86	2.90	3.31	2.20	4.35	4.25	3.09	3.59
C	2.38	5.69	2.50	2.72	3.14	2.00	4.28	2.48	1.78	2.98
E	2.86	3.14	2.32	2.04	3.88	1.26	4.08	1.98	2.48	2.61
G	0.50	6.36	1.90	3.90	3.28	1.10	4.05	1.38	1.18	2.63

SECOND DISTRIBUTION EXPERIMENT

SWATH B — BALES 3 and 4

Bale 3	Parts 1	2	3	4	5	6	7	8	9	mean
Sections	Percentage moisture content									
A	22.20	24.99	24.59	22.05	24.20	23.78	22.86	21.29	24.46	23.38
C	20.82	22.95	23.12	24.11	24.35	24.94	20.20	27.78	23.01	23.48
E	23.51	22.49	23.37	20.32	23.15	26.15	22.78	24.58	25.15	23.50
G	21.40	26.42	21.03	22.94	21.09	20.13	23.14	--	23.32	22.44
	Percentage propionic acid									
A	7.29	10.60	1.98	3.10	1.50	2.66	4.01	2.00	3.86	4.11
C	0.86	2.64	1.22	1.58	2.82	0.62	2.38	2.92	2.72	1.97
E	3.30	4.17	1.40	2.00	1.22	1.22	4.70	5.02	4.92	3.11
G	3.44	7.67	0.74	1.78	2.66	2.64	1.98	2.94	1.38	2,80

Bale 4	Parts 1	2	3	4	5	6	7	8	9	mean
Sections	Percentage moisture content									
A	23.69	26.01	24.10	26.78	25.53	25.50	22.45	22.72	25.08	24.65
C	22.34	24.87	22.85	24.60	23.77	26.12	24.75	25.03	28.55	25.10
E	23.39	25.33	24.26	27.30	22.88	23.26	24.50	25.82	26.13	24.76
G	23.18	24.53	25.24	23.84	25.62	24.83	24.94	23.45	26.69	24.70
	Percentage propionic acid									
A	2.72	3.19	1.10	1.74	2.46	1.14	2.92	4.29	4.10	2.56
C	8.38	10.90	9.70	5.55	4.24	6.67	6.39	6.50	12.93	7.92
E	3.55	6.28	5.79	2.86	1.82	1.70	4.39	3.76	3.60	3.75
G	2.12	8.11	1.86	4.09	3.30	1.38	3.88	2.50	1.74	3.22

SECOND DISTRIBUTION EXPERIMENT

SWATH C BALES 5 and 6

Bale 5	Parts 1	2	3	4	5	6	7	8	9	mean
Sections	Percentage moisture content									
A	29.39	31.08	32.03	25.09	30.04	32.37	22.68	26.23	30.28	28.80
C	25.00	33.39	-	22.03	30.53	30.09	29.96	29.81	30.44	28.90
E	25.27	30.87	29.82	31.81	29.08	28.24	27.99	28.32	31.36	29.13
G	27.85	38.42	37.54	30.16	32.63	30.95	28.72	24.31	24.80	30.59
	Percentage propionic acid									
A	1.42	3.10	1.86	2.00	4.71	0.70	1.14	2.40	0.74	2.01
C	1.62	3.93	2.12	3.97	5.13	6.53	-	2.64	2.22	3.62
E	1.50	2.54	0.82	1.26	3.45	1.26	1.10	1.98	1.82	1.76
G	2.96	5.12	1.98	2.34	3.11	3.00	2.54	3.89	2.40	3.04

Bale 6	Parts 1	2	3	4	5	6	7	8	9	mean
Sections	Percentage moisture content									
A	25.89	26.63	28.18	28.61	32.63	34.06	25.04	33.14	31.02	29.47
C	26.38	25.09	27.96	26.03	26.63	31.04	31.28	29.74	33.05	28.58
E	30.13	35.97	32.53	27.06	38.15	36.13	28.75	39.14	43.98	34.65
G	26.10	30.53	29.41	30.00	30.38	-	31.98	31.99	29.69	30.01
	Percentage propionic acid									
A	1.70	3.61	2.50	1.94	2.30	2.42	1.22	3.44	3.60	2.53
C	-	3.41	1.74	2.54	0.70	2.16	5.30	2.40	1.14	2.42
E	1.78	3.43	1.96	2.24	3.61	6.39	2.52	1.96	1.34	2.80
G	2.90	4.89	1.38	2.74	1.18	-	1.74	3.34	1.58	2.34

SECOND DISTRIBUTION EXPERIMENT

SWATH D — BALES 7 and 8

Bale 7	Parts 1	2	3	4	5	6	7	8	9	mean
Sections	Percentage moisture content									
A	24.79	25.27	26.39	-	28.50	28.54	29.08	27.36	28.26	27.27
C	24.39	26.38	24.89	26.01	24.27	24.88	26.24	27.03	24.67	25.41
E	-	28.95	34.50	27.46	28.61	31.93	23.21	26.64	27.30	28.58
G	25.88	28.41	25.11	25.65	27.01	24.94	22.28	23.56	27.19	25.56
	Percentage propionic acid									
A	3.14	3.51	2.32	-	3.51	2.88	1.38	0.90	3.81	2.68
C	1.94	5.58	2.32	1.90	1.62	4.10	-	2.76	3.90	3.02
E	0.86	4.28	4.80	0.98	2.46	2.36	1.66	1.38	3.10	2.43
G	2.56	5.82	2.86	3.89	1.62	1.94	3.56	1.90	-	3.02

Bale 8	Parts 1	2	3	4	5	6	7	8	9	mean
Sections	Percentage moisture content									
A	25.30	27.17	24.60	24.00	29.89	28.75	22.53	25.87	25.39	25.94
C	29.03	27.42	30.44	28.39	31.67	31.66	30.92	33.55	29.23	30.26
E	27.55	-	27.61	30.17	30.47	29.66	37.39	25.00	28.53	28.29
G	26.21	30.66	30.21	30.06	29.61	30.37	30.18	32.32	30.86	30.05
	Percentage propionic acid									
A	2.64	5.44	2.16	1.06	2.50	6.97	2.12	1.30	1.82	2.89
C	3.81	6.63	6.07	3.75	2.24	3.90	4.57	1.18	1.54	3.74
E	1.42	4.57	2.62	3.66	3.31	1.14	6.53	0.62	3.62	3.05
G	3.60	10.80	2.60	3.88	2.74	2.42	4.06	2.66	3.45	4.02

SECOND DISTRIBUTION EXPERIMENT

ANALYSES OF VARIANCE

Moisture Contents

	Even Moisture	Uneven Moisture	Total	Mean
Even Profile	23.23 24.04	28.41 30.58	106.26	26.57
Uneven Profile	23.56 24.60	26.89 28.62	103.77	25.94
Total	95.53	114.5	210.03	
Mean	23.88	28.63		26.25

Correction for the mean 5514.08

Analysis of variance table

Source	d.f.	Sum of Sqs.	Mean Sq.	Var. ratio
Moisture	1	44.98	44.98	37.17
Profile	1	0.77	0.77	
M. vs. P.	1	2.49	2.49	
Error	4	4.84	1.21	
Total	7	53.08		

Standard Error between means $= \sqrt{\frac{1.21}{2}} = 0.78$

Least significant difference = 0.78 x 2.78 = 2.16 (P = 0.05)

Analyses of the other results were carried out in exactly the same way and the following values were found :-

	Standard Error	L.S.D.
Mean propionic acid levels	0.28	0.78
Moisture co variance between sections	1.10	not sig.
Moisture co variance between parts	0.83	2.29
Acid level co variance between sections	3.54	9.84
Acid level co variance between parts	3.93	10.92

(P = 0.05)

SECOND DISTRIBUTION EXPERIMENT

STATISTICAL ANALYSES

Standard Deviations

	Moisture content		Propionic acid	
Bale	Parts	Sections	Parts	Sections
1	2.03	1.51	1.97	0.73
2	1.66	1.15	1.42	0.61
3	1.81	0.87	2.09	1.54
4	1.41	0.38	2.88	1.85
5	3.56	1.44	1.37	0.55
6	4.25	1.84	1.24	0.27
7	2.42	1.69	1.28	1.09
8	2.56	1.97	2.07	1.09

Coefficients of Variance

	Moisture content		Propionic acid	
Bale	Parts	Sections	Parts	Sections
1	8.54	6.51	47.18	17.25
2	6.69	4.79	48.04	19.40
3	7.78	3.67	69.71	42.88
4	5.70	1.53	65.21	48.90
5	12.11	5.07	53.77	23.21
6	13.86	6.02	48.57	10.71
7	9.10	6.30	46.22	37.03
8	8.96	6.89	60.34	38.82

APPENDIX 7.

Propionic acid Applicators

PROPIONIC ACID APPLICATORS

The three applicators used in the course of this experimental work were designed and supplied by BP Chemicals (International) Ltd., who also supplied technical help during their operation. In all the field experiments the same International B47 baler was used and the different applicators were fitted to it by the engineering department of the Edinburgh School of Agriculture Farms Division. The descriptions given below of the applicators and the notes on their use have been taken from the instructions supplied with the applicators by BP Chemicals (International) Ltd.

High Pressure Applicator (First Batch Experiment)

"The unit consists of a hydraulically operated piston pump which supplies acid to two horizontal spray bars, mounted across the pick-up chamber of the baler.

"Acid is drawn from a standard 5 gallon polythene container via stainless steel calibration vessel, to the pump. The dual head pump operates from the hydraulic system of the tractor and is designed to operate at a maximum working pressure of 600 pounds per square inch (p.s.i.). Since the tractor hydraulics are capable of supplying pressures well in excess of 600 p.s.i., a hydraulic pressure relief valve set at 750 p .s.i. has been placed across the oil inlet and outlet pipes. This valve should not require readjustment. In addition, a pressure relief valve has been placed over the acid delivery manifold. This has been preset to 470 ps.i. and should not be reset. In the event of excessive pressure being developed by the pump heads, which could cause damage, the valve will lift and allow the acid to be returned to the 5 gallon container. The valve also allows acid to be returned from the calibration vessel to the container during calibration operations.

"The hydraulic oil is passes to each side of the pump cylinder alternately via solenoid valves. The operating speed of the valve changeover, and hence the pump speed, is controlled by two electrical timers. The latter and the relay require a 12 volt supply taken from the tractor battery for operation, while the solenoid valves require 24 volts, taken from the tractor battery and an auxilliary battery in series.

"The pump is designed for use at a maximum rate of 100 strokes (cycles) per minute. This corresponds to 45 gallons per hour (g.p.h.) delivery rate. At the maximum flow rate the operating pressure is about 440 p.s.i., ie. slightly lower than the acid relief valve setting. At lower pumping rates, the operating pressures are correspondingly lower.

"Acid passes from the two pump heads into the manifold and thence to the two spray bars. A hydropneumatic accumulator is included in the manifold to stabilise the pump pulsations and pressure, which can be read on the pressure guage.

"The spray bars are positioned above and below the inlet to the hay intake chamber and each is fitted with an on/off valve and filter. Each spray bar has 18 x 0.0125" diameter holes giving a maximum of 36 acid application jets. Provision has been made for the blanking off of some of the holes with jubilee clips to maintain the pressures at lower pumping rates."

Treatment of hay

a. Bale a short length of swath and from the weight of the bales calculate the rate of baling.

b. Calculate the rate of acid application required to give the intended

level of acid treatment to the hay

c. With reference to the calibration graph supplied by BP Chemicals*, set the timers to the correct number of strokes p r minute in order to supply the required rate of acid.

d. Open both spray bar valves.

e. Open oil inlet valve and commence baling at the pre-determined rate.

d. When baling is completed, or during any breaks in baling, close the oil inlet valve and switch off electrical supplies.

(* The calibration data was checked before commencing the experimental work.)

Low Pressure Applicator, Mark I (Second batch experiment and First Distribution experiment)

"The low pressure applicator has been designed to apply propionic acid to hay while baling is in progress. The system comprises a Beresford PV31 centrifugal pump powered by a 240 volt electric motor. A valuable feature of this type of pump is that it can safely be run with the delivery line completely closed. However, it is not recommended that this should be done for extended periods of time. The power supply for the motor is by a 'Dynamotor' operating off the 12 volt tractor battery.

"The spray nozzles are used in pairs and have been calibrated with propionic acid to give a range of acid throughputs. The maximum and minimum acid throughput values are quoted for each pair of nozzles. The minimum value was taken as each of the nozzles gave a 30 inch spread of acid when fitted at a vertical height of 21 inches above the baler floor (total spread for both nozzles, 60 inches).

"This enables one pair of nozzles to be used for a range of

acid treatment levels and also ensures that the whole of the baler pick-up area is covered by the acid spray.

"The pipe work for the acid supply is constructed of steel with fittings in 0.5 and 0.25 inch B.S.P. The flow control valves and stopcocks are of stainless steel. The flexible pipe is of polythene. The acid supply resevoir comprises a 5 gallon polythene container held to the roof of the baler by a 'Dexion' frame."

Operating instructions

a. Prime the pump using water (for safety reasons).

b. Calibrate the baling speed by running the baler at the speed at which it is going to be used during the experiment and weighing the bales produced in a known time.

c. Select the nozzle pair to give the required acid treatment level range. If the nozzles are not going to be used at the maximum

Nozzle pair	Max. flow rate	Min. flow rate
J 100	32.11 g.p.h.	14.53 g.p.h.
K 100	46.00	17.00
L 100	62.65	31.02
N 100	80.00	35.50

flow rate then they must be set and calibrated to give the flow rate required.

d. Switch on the pump, open the stop cock and commence baling at the pre arranged rate.

e. At the end of each run, and when changing acid containers switch off the acid at the stop cock and so prevent having to re-prime the system.

f. At the end of operations drain the system and wash out with water.

Low Pressure Applicator, Mark II (Second distribution experiment)

A number of madifications were made to the design of the low pressure applicator in order to simplify its operation and improve its efficiency in distributing the acid evenly through the bales.

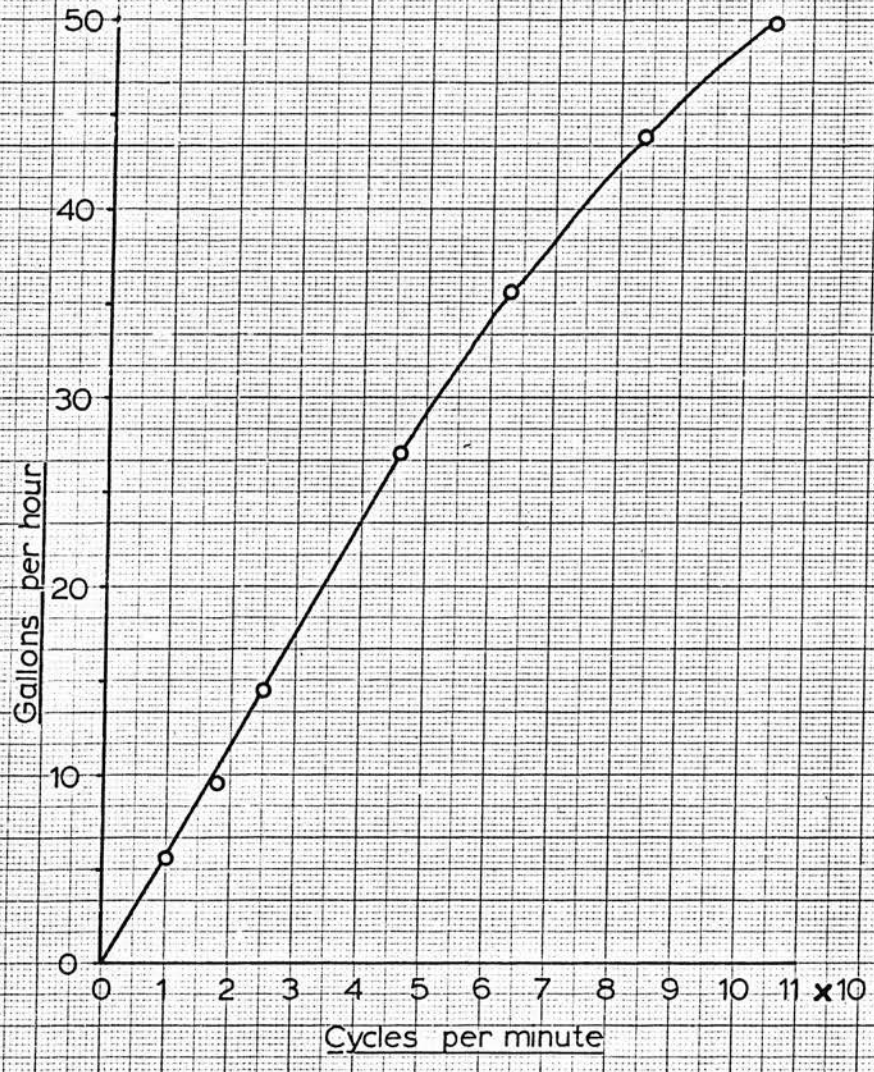
a. The 5 gallon polythene container for the acid was fitted to a hinged frame so that as the container was fitted in place it was inverted. The whole system then automatically became primed with acid as soon as the valves were opened.

b. A flow meter was built into the system so that the rate of acid flow could be measured and adjusted at any time.

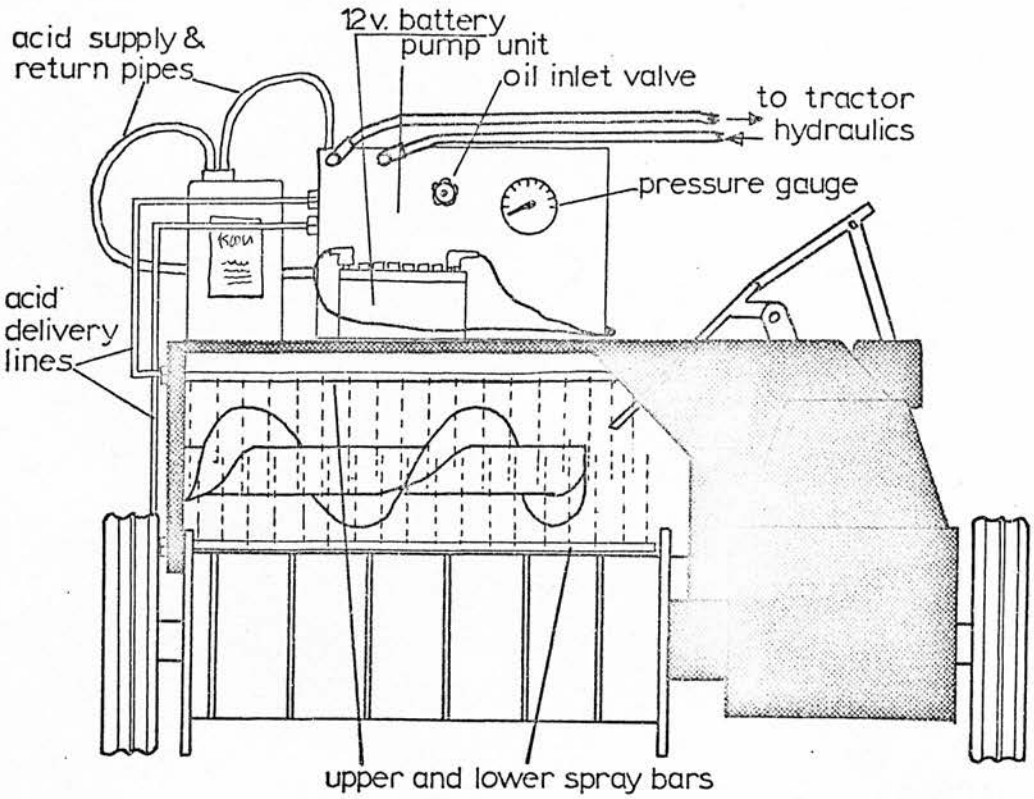
c. The position of the acid nozzles was altered from above the pick-up reel of the baler to across the inlet to the bale chamber. The nozzles were located on opposite sides of the inlet, but not exactly opposite one another so that they produced two parallel curtains of acid which overlapped, but did not collide.

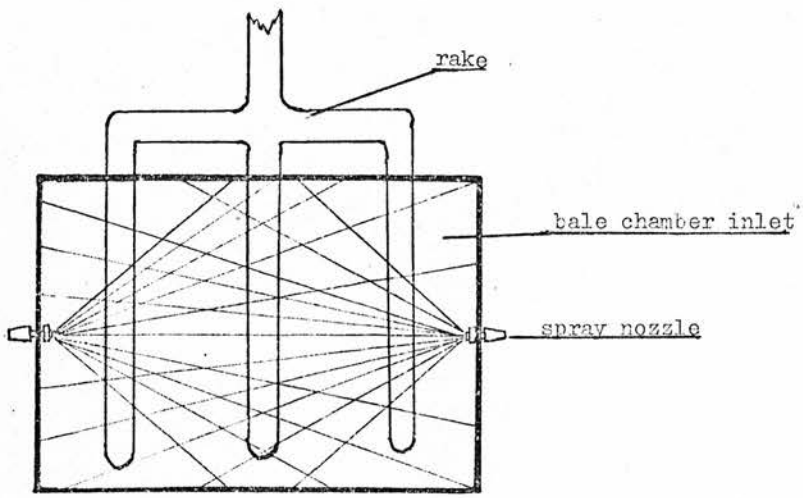
CALIBRATION DATA FOR HIGH PRESSURE APPLICATOR

Supplied by B.P. Chemicals (International) Ltd.



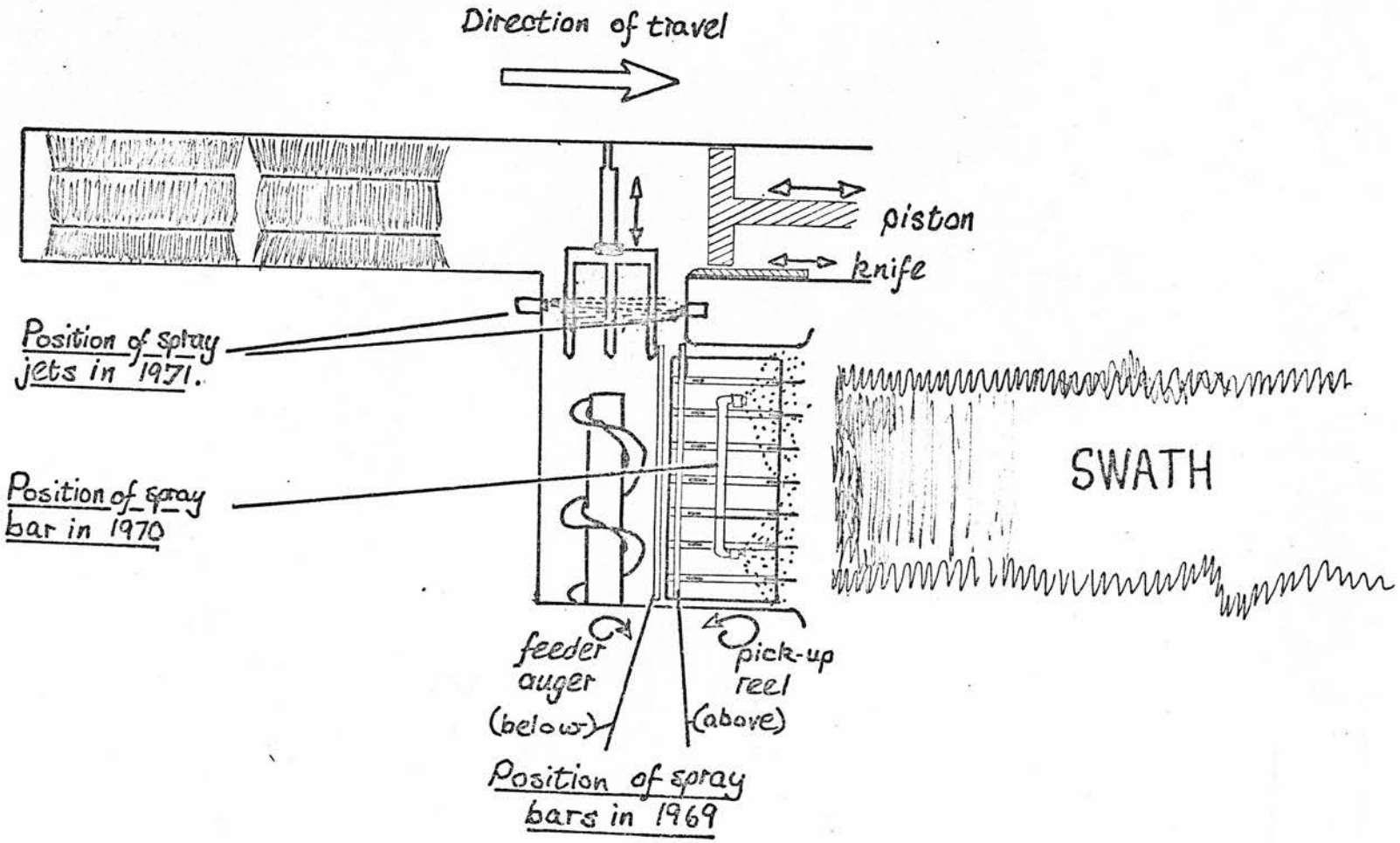
HIGH PRESSURE APPLICATOR





LOW PRESSURE MARK II APPLICATOR POSITION OF SPRAY NOZZLES

PLAN OF BALER SHOWING POSITION OF SPRAY JETS IN 1969, 1970 and 1971



APPENDIX 8.

'Quick Drier' for Moisture Determinations

'QUICK DRIER' FOR MOISTURE DETERMINATIONS

The drier consists of a hot air blower unit, a specially constructed air duct and two mesh bottomed trays which fit onto the top of the air duct. The blower unit was made from an existing unit, manufactured by Tower Silos Ltd., from which the 1 kilowatt heater element was removed and replaced by a 2 kilowatt element. The wire gauze over the top of the blower unit was also removed. The air duct was made so that it would fit neatly into the top of the blower unit.

Moisture Determinations

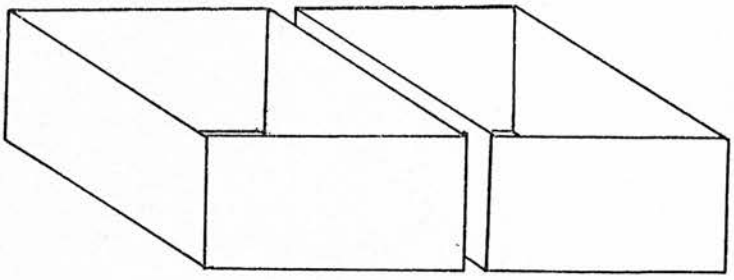
The sample of hay is chopped into 1 cm lengths and thoroughly mixed. Fifty or 100 grammes of the sample are weighed into each of the trays which are then placed on the drier. The bigger sample weight is used when the moisture content is expected to be over about 50 per cent. Samples with a low moisture content dry to a constant weight in about 30 minutes whereas 45 minutes are needed at the higher moisture levels.

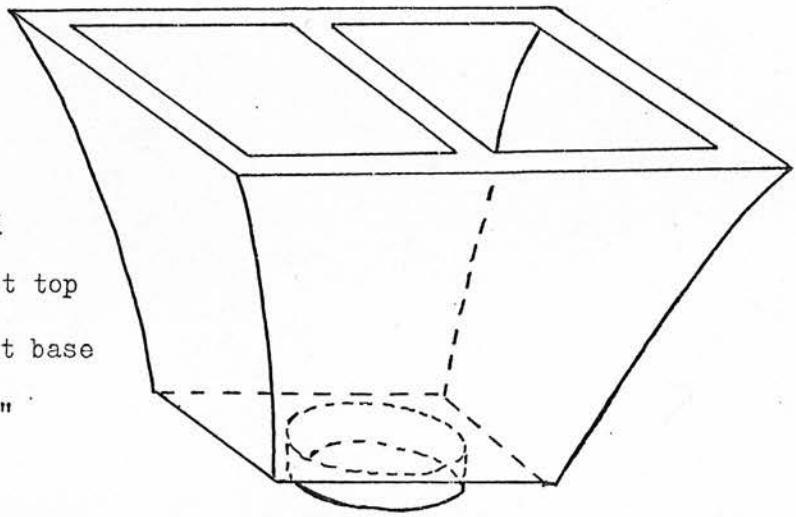
Comparison with Oven Determinations

The results in the table on the next page were obtained from samples for which both oven and 'quick drier' determinations were made. All the results are means of duplicate determinations. The results for the quick drier are within $\pm$ 3 per cent moisture of the oven determinations. Although the mean of the quick drier determinations is 0.9 per cent below that of the oven determinations, this difference is not significant even at $P = 0.25$.

'QUICK DRIER' FOR MOISTURE DETERMINATIONS

Two mesh bottomed sample trays 9" x 9" x 6"



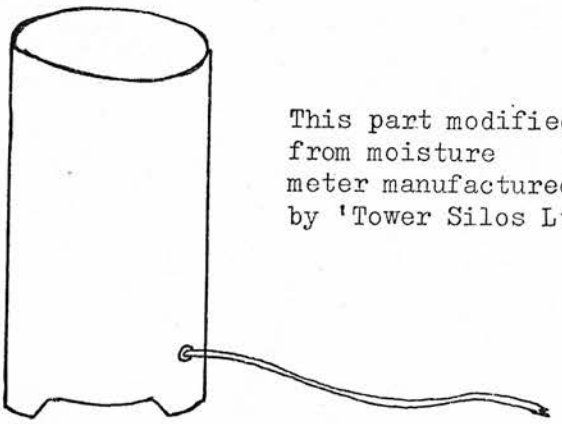


Air duct

20" x 11" at top

9" x 9" at base

height 9 "



Blower unit with 2 kilowatt element and an electric fan

This part modified from moisture meter manufactured by 'Tower Silos Ltd.'

Comparison of Moisture Determinations made using the 'Quick Drier' and Oven drying

Percent moisture content

	Oven drier	Quick drier	Difference
	69.4	69.9	+0.8
	66.2	63.6	-2.6
	64.2	64.0	-0.2
	51.9	50.0	-1.9
	50.5	49.2	-1.3
	49.0	48.1	-0.9
	48.6	49.6	+1.0
	44.8	43.6	-1.2
	34.6	35.1	+0.5
	33.6	30.8	-2.8
Mean	51.3	50.4	-0.9

APPENDIX 9.

The Method of Mini-bale Production

THE METHOD OF MINI-BALE PRODUCTION

Reference should be made to the diagrams of the mini--balers which are given in figure 28 and to the photographs of the Mark II Mini-baler which are on page 156. The procedure given below is described as for the Mark II Mini-baler, but is essentially the same as that used when making bales with the Mark I model.

1. With the hydraulic jack lowered so that the sliding base is at the bottom of the bale chamber, insert a length of baler twine through each of the slits in one side of the bale chamber and across and out of the slits in the opposite side.
2. Fill the bale chamber with the hay.
3. Taking each of the two strings on one side of the baler in turn, feed the string back through the slit, over the top of the hay, and out through the opposite slit. All four ends of the strings should now be on the same side.
4. Close and secure the lid.
5. Pump up the hydraulic jack so that the sliding base is pushed up and the hay compressed to the required density. It may be necessary to adjust the screw part of the jack in order to give the desired degree of compression.
6. Tie the strings in the two slits as tightly as possible.
7. Release the catch, open the lid and remove the mini-bale.
8. Trim off any loose strands of hay and excess string from the bale before weighing.

APPENDIX 10

Methods for Chemical Analyses

Determination of Water Soluble Carbohydrates in Hay

Extraction The sample of hay from the bale or swath is chopped into 1 inch lengths and 2 samples of 10* grammes each are weighed out. The dry matter of the sample is determined on the remainder. The 15 g. samples are macerated with 250 ml chilled water in an 'Osterizer' for 5 minutes. The suspension is filtered through linen and a 100 ml aliquot of the filtrate is removed. The pH value of the sample is determined on the remaining filtrate using a Pye Universal pH Meter. The 100 ml aliquot is acidified with 0.1 $\overline{N}$ H_2SO_4, boiled and allowed to cool before it is filtered through Whatman No.1 paper. The residue and filter paper are washed thoroughly with deionised water and the filtrate and washings are made up to 250 ml.

Hydrolysis Five ml. of 2 $\overline{N}$ H_2SO_4 are added to 15 ml of this solution in a boiling tube. The tube is placed in a boiling water bath for 10 minutes, cooled and the solution neutralized to methyl red and made up to 100 ml. The water soluble carbohydrates are determined on 5 ml. of this solution using the modified Somogyi reagent.

Modified Somogyi Reagent Fifty six grammes of di-sodium phosphate and 40 g of potassium sodium tartrate are dissolved in about 700 ml of distilled water, 3.3 g sodium hydroxide dissolved in 100 ml deionized water are added and then, with stirring, 80 ml of 10 per cent copper sulphate ($CuSO_4.5H_2O$) solution are introduced. Finally 180 g of anhydrous sodium sulphate are added and dissolved and the solution is diluted to 1 litre. This reagent is kept at a temperature above 20°C to prevent crystallisation.

Twenty five milli-litres of 0.1 $\overline{N}$ potassium iodate per litre of Somogyi reagent are then added. This concentration is suitable

for detecting up to 3 mg of sugar in 5 ml of solution.

Method Five milli-litres of Somogyi reagent are added to 5 ml of sugar solution in a boiling tube. The tube is placed in a boiling water bath for 20 minutes, and cooled for 5 minutes. Two milli-litres of 2.5 per cent potassium iodide are added down the side of the tube, followed by 2.5 ml of 2 $\overline{N}$ H_2SO_4. The contents of the tube are shaken for 10 seconds and titrated with 0.005 $\overline{N}$ sodium thiosulphate using starch as the indicator.

* At moisture contents of above about 50 per cent a 25 g sample was used, macerated with 200 ml of water.

PROPIONIC ACID DETERMINATION ON HAY

Titration Method

Preliminary investigations into this method of determining the level of propionic acid on hay were carried out on the hay from the second mini-bale experiment which had been treated with known levels of propionic acid. Chopped samples of the hay, 10 g was found to be a convenient size, were placed in polythene bags along with 300 ml of deionised water and the bag sealed.

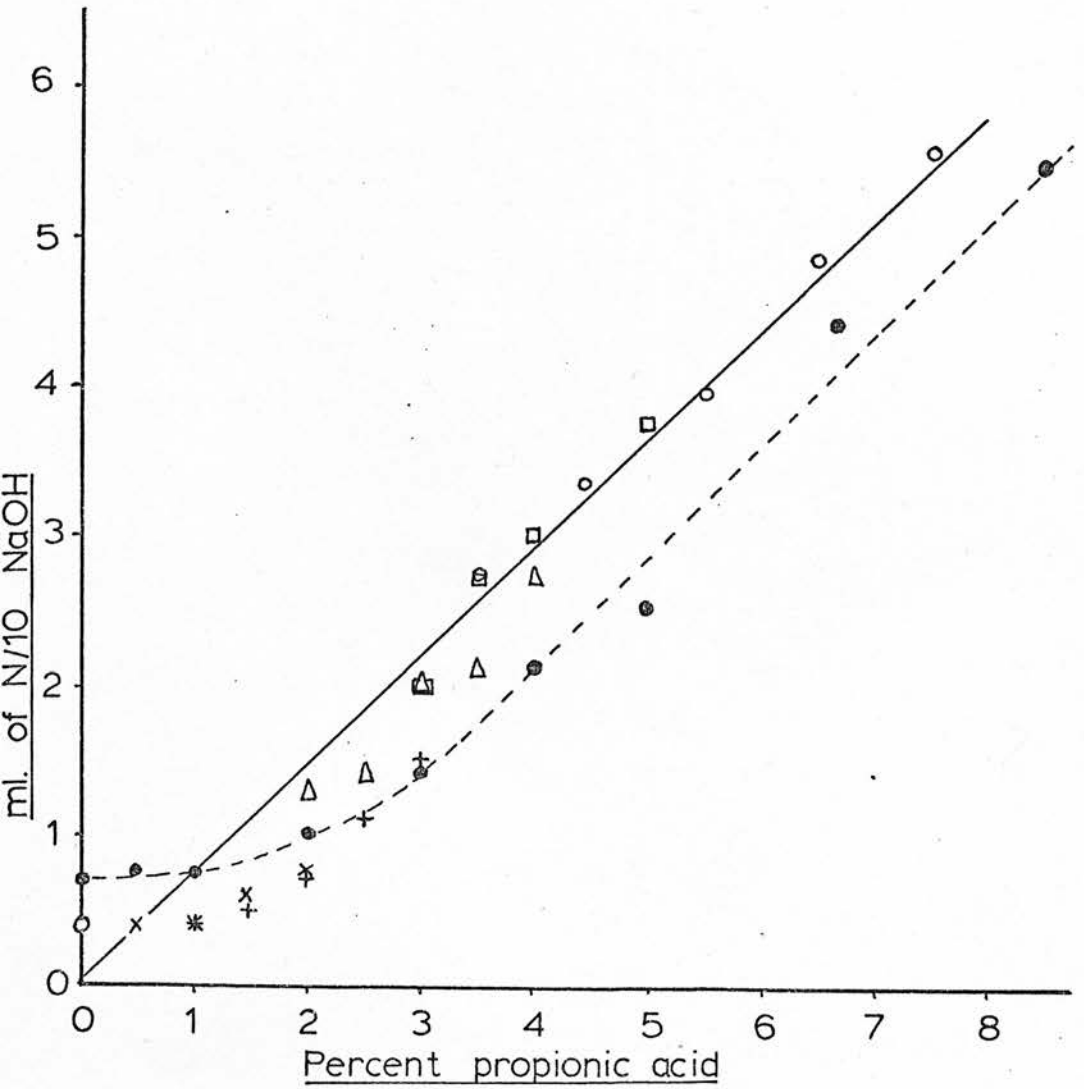
After being left to soak overnight at room temperature the corner of the bag was cut off and the solution collected in a beaker. The sampling had to be carried out within 24 hours otherwise the contents of the bag were liable to the growth of bacteria.

25 ml. aliquots of the solution were titrated with N/10 sodium hydroxide solution using phenolpthalene indicator. The reults of the titrations are given on the graph on the next page. In the hays at 76 and 60 per cent moisture the recovery of propionic acid was virtually 100 per cent for levels of acid between 4 and 8 per cent. On the lower moisture hays, however, the recovery of the propionic acid was less than 100 per cent. This was also found to be the case in the standards prepared for the second distribution experiment in which the hay had a moisture content of about 30 per cent.

There would seem to be two possible reasons for this lower recovery of acid from the low moisture hays. It may be that some of the acid reacts with components of the hay to form insoluble compounds which could not be recovered during the extraction procedure.

PROPIONIC ACID DETERMINATION ON HAY

Titration Method



○ 76 per cent moisture level
□ 60 per cent moisture level
△ 50 per cent moisture level
\+ 40 per cent moisture level
× 30 per cent moisture level

} Second mini-bale experiment

--●-- Standards from second distribution experiment

——— Theoretical 100 per cent recovery of acid

Alternatively it is possible that during the treatment of the hay with the propionic acid the drier hay took longer to absorb the acid. The surface of the hay would then remain 'wet' with the acid for a short time and would be liable to loose acid by volatilization. According to the results of the standards for the second distribution experiment the recover of the acid ranged from about 65 per cent at the lower acid levels to 85 per cent at the higher levels.

Standards for Second Distribution Experiment

Hay from the experimental swaths was brought inside and treated with propionic acid at 0.5, 1.0, 2.0, 3.0, 4.0, and 5.0 per cent by weight using a hand sprayer. These hay samples were used to obtain standard titrations at the start of the experiment and were also used periodically to check for any subsequent changes during storage. The hay standards were kept along with the other samples in the deep freeze. The following table shows the results of the repeat analyses and the mean values which were used for plotting the graph.

	nil	0.5	1.0	2.0	3.0	4.0	5.0
Initial (25th July)	0.93	0.68	0.72	1.08	1.48	2.17	2.67
9th August	-	-	0.80	-	-	-	2.58
15th August	-	-	0.77	-	-	-	2.47
7th September	0.54	0.60	0.67	0.93	1.34	2.00	2.32
22nd September	0.56	0.97	0.80	1.01	-	-	2.47
29th September	0.85	0.81	0.73	1.02	1.40	2.10	2.49
Mean	0.72	0.77	0.75	1.01	1.41	2.09	2.50

During the experiment it was found that a number of the parts of the bales had received more than 5 per cent acid and so

a number of the original standards were retreated so that the acid levels were brought up to 6, 7, 8.5 and 10 per cent. These retreated samples were then used to obtain standard titrations for the acid levels up to 10 per cent with the following results:-

Per cent acid	5.0	6.0	7.0	8.5	10.0
Titration	2.48	4.49	4.22	5.43	6.20

Even the 10 per cent acid level was not found to be high enough for some of the experimental samples, but in this case the graph (Figure 46) was extrapolated to 13 per cent acid to provide an acid level estimate for these samples.

APPENDIX 11.

AIR DISPLACEMENT SAMPLER

Results from preliminary experiment

Two hays were selected, one which had no visible moulding and one which was showing severe moulding. The counts were obtained using the air displacement sampler as outlined in the first batch experiment. The glass jar and filter funnel were sterilized between samples (Figure 7). Two replicates were made for each plate.

	Moulds		Bacteria and actinomycetes		
	Malt salt agar		Nutrient agar		
	27°C	36°C	27°C	36°C	55°C
Mould free sample	16	4	530	536	50
	8	10	79	950	39
Mouldy sample	54	20	1600	1500	640
	36	28	1200	1500	37

Comparison of results from air displacement and serial dilution methods.

Two of the samples for which the results of counts obtained using the air displacement sampler are given in table 11 were also used to obtain counts by the serial dilution method. The results are given below.

	Moulds		Bacteria and Actinomycetes		
	Malt salt agar		Nutrient Agar		
	27°C	36°C	27°C	36°C	55°C
Sample 2 (Mould free)					
Air displacement	5	5	20	6	5
	7	6	79	17	21
Serial dilution	43	22	5400	490	85
Sample 6 (Mouldy)					
Air displacement	4	13	236	127	2000
	45	54	332	480	3000
Serial dilution	4,700	13,000	1,200,000	1,390,000	690,000

APPENDIX 12

SERIAL DILUTION TECHNIQUE

Five grams of the chopped hay sample are placed in a top drive macerator along with 495 ml of sterile water for 2 minutes. From this 1 in 100 dilution, further dilutions down to 1 in 10^7 are made by pipetting 1 ml of each dilution into 9 ml of sterilized water. From each dilution from which a count is to be taken 1 ml is pipetted into a sterile petri dish and approximately 5 ml of the appropriate agar at a temperature of about $45^{\circ}C$ poured in. The contents of the petri dish are gently mixed and then allowed to set.

After incubation counts are taken at three or five days, depending upon the speed at which the colonies are developing.

APPENDIX 13

COMPARISON OF BALE DENSITIES

Grammes per cubic centimetre

	Fresh matter density	Dry matter density
First Batch Experiment		
Low moisture level	.1755	.1279
Medium moisture level	.2024	.1303
High moisture level	.2306	.1305
First Mini-bale Experiment		
50 per cent moisture	.170	.085
Second Mini-bale Experiment		
76 per cent moisture	.381	.091
60 per cent moisture	.272	.109
50 per cent moisture	.204	.102
40 per cent moisture	.170	.102
30 per cent moisture	.150	.105

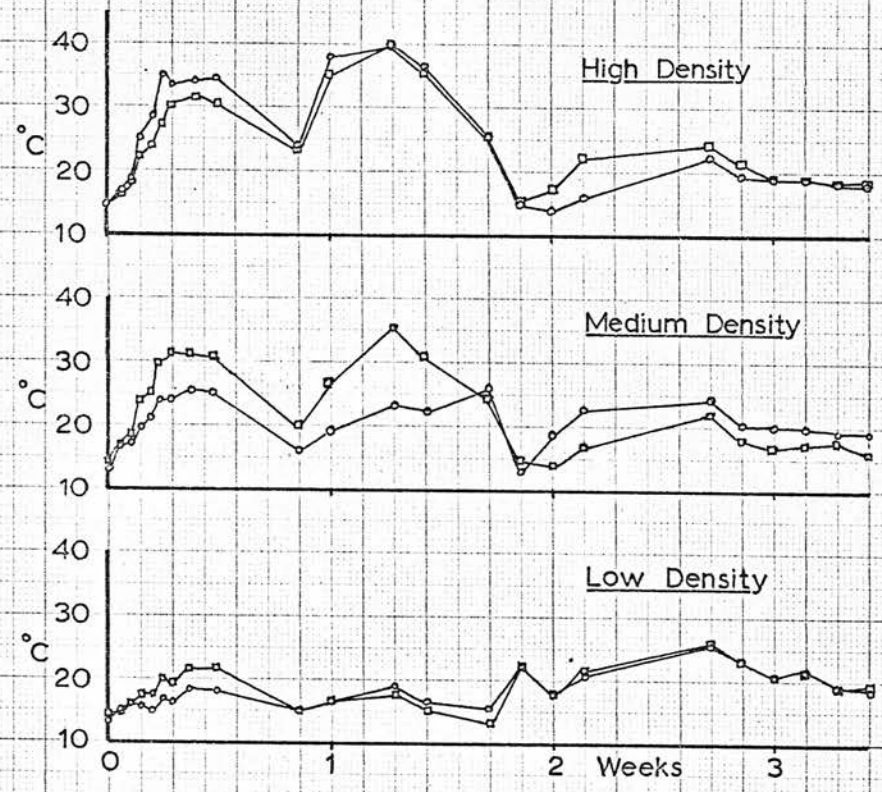
MINI - BALE DENSITY EXPERIMENT

Two mini-bales were made at each of three densities using hay with a moisture content of about 47 per cent. For the highest density 2000 g of the hay were placed in the Mark II mini-baler and the bales made at the highest compression that the baler could achieve. Bales at the two lower densities were made by using 1500g and 1000g and compressing the bales to the same size as the highest density bales. The densities were as follows:-

	Fresh matter density	Dry matter density
High density	.272	.144
Medium density	.204	.108
Low density	.136	.072

FIGURE

EFFECT OF DENSITY ON TEMPERATURE
IN MINI - BALES AT 47 PER CENT MOISTURE



The bales were stored in insulated boxes as in the other mini-bale experiments, and thermometers were inserted through the lids so that the temperatures could be read at regular intervals.

Results

The temperature patterns of the bales at the three densities over the first 3 weeks of storage are shown in the figure. By the end of the 3 week period all the bales were completely mouldy.

The results show clearly that there is a tendency for the temperature in the bales to increase as the density increases. The high density bales were more dense than any of the bales in the other mini-bale experiments and had temperatures slightly above those in the untreated bales at about 50 per cent moisture in those experiments. The high density bales were also more dense than the high moisture bales in the first batch experiment. The conventional sized bales, however, reached temperatures of over 60°C (Figure 11), whereas the mini-bales barely reached 40°C. It is likely, therefore, that the larger surface to volume ratio of the mini-bales led to a more rapid rate of heat loss in the mini-bales in spite of the polystyrene insulation surrounding them.

The temperatures in the medium moisture mini-bales were similar to those in the untreated bales in the other mini-bale experiments at the equivalent moisture levels. In the low density mini-bales, however, the temperature rises were slight. The density of these bales was below that of any of the other mini-bales or conventional sized bales in the other experiments.

The results of this experiment are in accord with those of Nelson (1966, 1968 and 1972), who carried out similar experiments using full sized bales.